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THE POSSIBILITY OF COMMERCIAL PRODUCTION OF BUTYRIC ACID FROM RAW SUGAR AND WASTE MOLASSES [1]

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Butyric acid is an important chemical used in many organic syntheses and in some industries. It is extensively used in the leather industry as an excellent softener, in the manufacture of plasticizers, in lacquer making, as a component of drugs, esters, dryers, varnish, cosmetics, and flavoring for rum.

Commercial production of butyric acid has gained popularity only very recently, although its production by bacteria has been recognized since the time of Pasteur. Only within very recent years have efficient fermentation processes for producing the acid as a major product been developed.

Production of butyric acid has proved to be a most valuable adjunct to some of the large producers of industrial alcohol from molasses. The residual sugar in the alcohol distilleries can be utilized profitably in the production of butyric acid.

If butyric acid can be produced profitably on a commercial scale from molasses and raw sugar, excess sugar and waste molasses of the Philippine sugar industry could be disposed of easily. This will mean a greater acreage of sugar cane which will not necessarily affect the sugar quota for export and domestic consumption. Commercial fermentations for the production of butyric acid from sugar and molasses may thus partly solve our sugar problem. Hence a study was made to determine the conditions favorable for butyric acid production from raw cane sugar and waste molasses by bacterial processes.

Review of literature

Studies on the nutritional requirements of butyric acid-forming bacteria on glucose and other fermentable sugars were conducted by Baier, and Büchner and Meisenheimer (Owen, 1936), Buchanan

[1] Experiment Station contribution No. 1432.

and Buchanan (1913), Le Franc (1914, 1927), Hibbert (1918), Arroyo (1934), and Owen (1936).

Waksman and Stevens (1930), working on butyric acid and butyl alcohol fermentation of hemicelluloses and starches found that the *Clostridium butyricum* Prazmowski group differed from the butyric acid bacteria in that this group grew not only on sugars and starches but also on certain hemicellulosic materials.

Work on the recovery and quantitative determination of butyric acid from fermenting mashes were undertaken by Harden (1901), Olmstead, Whitaker and Duden (1929), and Klinc (1931).

MATERIALS AND METHODS

Apparatus and reagents. The apparatus and reagents used in this study were those generally used in chemical and sugar laboratories. Most of the solutions and reagents were prepared in accordance with directions in the Official and tentative methods of analysis of the Association of Official Agricultural Chemists (1935).

Source of the organism used. The organism was isolated from annato seeds (*Bixa orellana* Linn.) and grown on potato-dextrose agar. A liquid medium containing annato seed extract, distilled water, and sodium carbonate was also used.

Molasses and raw cane sugar. The molasses and raw cane sugar were from the 1939 and 1940 crops of the U. P. Sugar Mill.

The acidifier. About fifty annato seeds were placed in a test tube of distilled water made slightly alkaline with sodium carbonate and incubated at 31°C. After three or four days, rapid fermentation was observed and the scent of butyric acid was very noticeable. The solution turned from slightly alkaline to acid in reaction. The color of the solution changed from light red to yellowish.

Molasses and sugar solutions for inoculation

Preliminary runs

Molasses solution. In the preliminary experiments, an 11 per cent molasses solution was used. Calcium carbonate was added gradually at the rate of 10, 15, 20, and 25 grams per 100 grams of solution. Three samples were used for each treatment. All fermentations were carried out in flasks plugged with cotton. The solution was sterilized in an autoclave at ten pounds pressure for thirty minutes. The sterilized culture solution was inoculated with

the actively growing cultures and then incubated for two weeks at 30°C. After fermentation, the fermented mash was analyzed for sucrose, glucose, and butyric acid.

Sugar solution. Preliminary runs were made to determine the right amount of calcium carbonate to add to the culture solution. A ten per cent sugar solution was used. The amounts of calcium carbonate added per 100 grams sugar solution were 4, 8, 12, and 16 grams for each treatment. The same procedure was followed in the case of the molasses solution. Because of the limited capacity of the incubator, the runs were made in triplicate only.

Final runs

Molasses solution. Fifteen grams of calcium carbonate (the best in the preliminary trials) were added per 100 grams of molasses solution. The concentrations used were 8°, 10°, 12°, 14°, 16°, 18°, and 20° Brix. For each concentration, three samples were used. They were placed in flasks plugged with cotton and sterilized at ten pounds pressure for thirty minutes. Inoculation of the culture solution was done with a platinum wire loop. The wire loop was flamed, dipped in sterile water, and then inserted in the test tube containing the pure culture of the organism. A little portion of the dextrose-agar medium was taken out and inserted in one of the culture media.

Sugar solution. Four grams of calcium carbonate were added to the sugar solution per 100 grams sample. The solutions used were also 8°, 10°, 12°, 14°, 16°, 18°, and 20° Brix. The solution was prepared and inoculated with the pure culture of the organism, similar to that of the molasses.

Runs on both molasses and sugar solutions were for a period of only three weeks. As shown by the data on butyric acid production, the fermentation could not be carried on profitably beyond three weeks.

Recovery of butyric acid from fermented mash. The fermented liquor was neutralized with calcium hydroxide and evaporated to approximately one-fourth its original volume. The evaporated liquor was cooled and then acidified with concentrated sulfuric acid to liberate the butyric acid from its salt. The acidified fermented liquor was then filtered, and the filtrate used for butyric acid determination.

Temperature of incubation. An improvised incubator maintained at a temperature of 31°C was used. Because of the limited capacity of the incubator, only a few replications were possible.

Analyses of molasses and sugar solutions, fermenting mash, and sulfuric acid-treated mash: Sucrose. The sucrose and glucose in the original sugar and molasses solutions were determined by Walker's inversion, and Lane and Eynon's methylene blue methods respectively as described in the Methods of chemical control for cane sugar factories of the Association of Hawaiian Sugar Technologists (1931).

The total original invert sugars were obtained by adding the glucose content of the original sample and the sucrose value multiplied by 1.05. The same procedure was employed in the determination of invert sugars in the fermented mash.

Glucose. In the determination of glucose proper dilution was made in such a way that 15 to 50 ml. of the clarified solution were needed to react with 10 ml. of standard Soxhlet's solution. For sugar solutions, a dilution was made in such a way that the sucrose content did not exceed 5 grams per 100 ml. of the prepared solution, and for the molasses solution, 1 gram per 100 ml. This method of determination was employed both in the original sugar and molasses solutions and in the fermented liquors.

Butyric acid. In order to determine qualitatively if butyric acid was produced, an aliquot portion of the acidified mash was steam-distilled. A small portion of the distillate was neutralized and evaporated to dryness on a steam bath. The residue obtained was treated with five ml. of 10 per cent sulfuric acid. The characteristic odor of butyric acid evolved was noticed.

Rectification of butyric acid from the sulfuric acid-treated mash. The acidified mash was steam-distilled, and enough distillate was collected in an Erlenmeyer flask. The butyric acid was recovered from the distillate by the ether extraction method of Harden (1901) as follows: The distillate was placed in a separatory funnel, and ether was added. The mixture was shaken thoroughly before the ethereal layer was separated from the aqueous solution. The aqueous solution was further extracted with ether to recover the residual acid from the previous extraction. This process was repeated three to four times to insure complete extraction of the butyric acid. The combined extracts, after part of the ether had been recovered, was titrated with 0.1N sodium hydroxide solution. The weight of bu-

tyric acid recovered from the acidified mash was calculated from the volume and normality of the alkali used.

RESULTS AND DISCUSSIONS

The results obtained in these experiments are presented in tables 1 to 4.

Butyric acid production in molasses solutions

Table 1 shows the influence of varying amounts of calcium carbonate added per 100 grams molasses on an 11 per cent solution. The butyric acid contents were calculated on the basis of the sugars consumed, and on total initial sugars. The 15-gram treatment gave the highest per cent yield of butyric acid (21.78 ± 0.53 per cent based on sugars consumed, and 17.49 ± 0.47 per cent based on total initial sugars). Molasses solutions without calcium carbonate gave low yields of butyric acid. The 20- and 25-gram treatments also gave low yields, similar to that of the control. Buchanan and Buchanan (1920) state that butyric acid production in sugar solution in the absence of calcium carbonate is inhibited by an increase in the acidity of the mash.

The superiority of the 15-gram treatment over the others, including the control, is shown in table 2. The yield from the 15-gram treatment was significantly greater than that from any of the other treatments. In economy (butyric acid based on sugars consumed) and efficiency (butyric acid based on total initial sugars) of butyric acid production on an 11 per cent molasses solution, the 15-gram calcium carbonate treatment was the best.

Table 3 shows the effect of sugar concentration of molasses solutions on the yield of butyric acid. A 12° Brix molasses solution gave the best yield of butyric acid (on the basis of sugars consumed and on total initial sugars). The average yields of butyric acid from the first to the third week was 35.86 ± 1.08, 23.85 ± 0.60, and 20.31 ± 0.37 per cent, respectively, based on sugars consumed; and 12.81 ± 0.20, 13.78 ± 0.24 and 14.95 ± 0.26 per cent, respectively, on the basis of total initial sugars. Values on economy of butyric acid production at this concentration decreased as expected, up to the end of the third week, when final analysis was made. Values on efficiency of butyric acid production increased from the first to the third week with a maximum value of 14.95 ± 0.26 per cent. Although high yields of butyric acid were also obtained from the 8° and 10° Brix molasses solutions,

these when statistically compared with those obtained from the 12° Brix solution were very significantly inferior (table 4), except in the third week, where insignificant differences were obtained. The sugar concentration in the 12° Brix molasses solution was found to be 0.8515 grams per 10 ml. (approximately 8.52 per cent). Arroyo (1934) found that the best yield of butyric acid on blackstrap molasses ranged from 46 to 48.5 per cent. Considering the length of incubation period in point of economy, one week was the best. However, the most economical period of incubation did not give the highest per cent of butyric acid.

The 12° Brix molasses solution gave the highest per cent yield of butyric acid, so it was selected as the basis of comparison in treating the data statistically (table 4). Significant differences were obtained when 12° Brix was compared with all concentrations, in favor of the 12° Brix, except with 8° and 10° Brix during the third week. This indicates that for the production of butyric acid, 8° as well as 10° Brix may be employed as efficiently as 12° Brix molasses solution.

Production of butyric acid in sugar solutions

Table 1 shows the influence of varying amounts of calcium carbonate added per 100 grams of a 10 per cent raw sugar solution on the production of butyric acid. In table 1 the 4-gram treatment gave the highest per cent yield of butyric acid, with an average of 31.08 ± 0.51 per cent based on sugars consumed and 9.58 ± 0.16 per cent based on total initial sugars, whereas the 12-gram treatment gave the lowest yield of butyric acid with an average of 14.63 ± 0.38 per cent based on sugars consumed and 4.46 ± 0.11 per cent based on initial sugars. The 8-, 12-, and 16-gram treatments and the control gave low yields of butyric acid.

The superiority of the 4-gram treatment over the others, including control, is shown in table 2. The 4-gram $CaCO_3$ treatment was selected as the basis of comparison, for it was found in the preliminary tests to be the best in the production of butyric acid on a 10 per cent sugar solution. Highly significant differences were obtained in favor of the 4-gram treatment both in points of economy and efficiency. The addition of 4 grams calcium carbonate per 100 grams sample tended to increase sugar utilization but the addition of excess calcium carbonate had no effect upon acid production.

The effects of sugar concentrations on the production of butyric acid are presented in table 3. The 12° Brix sugar soltuion gave

the highest yield of butyric acid after the first week with an average of 56.67 ± 2.07 per cent based on sugars consumed (economy). The yield of butyric acid decreased up to the end of the third week with 20.52 ± 0.53 per cent. In point of efficiency, 18° and 20° Brix were found unfavorable, particularly after the second and third weeks of incubation. The 18° and 20° Brix sugar solutions gave consistently lower yields of butyric acid than the others. An optimum efficiency was obtained in the tests from 8° Brix sugar solution, with an average of 14.16 ± 0.67 per cent of butyric acid after the third week, based on the initial sugars.

Table 4 gives the comparative influences of varying sugar concentrations on the production of butyric acid; 8° Brix sugar solution was taken as the basis of comparison. As shown in table 4, insignificant differences, in point of economy, after the third week were obtained when the yield from the 8° Brix sugar solution was compared with the others, except in the case of the 14° Brix solution, where the difference was only slightly significant in favor of the 8° Brix solution. This single exception may be attributed to the relatively low variation of the individual runs, as shown by the low probable error of the mean from the 14° Brix sugar solution. On the whole, in point of economy of butyric acid production, the different sugar concentration were all the same.

A different situation, however, was obtained when efficiency of butyric acid production was considered. Significant differences, in favor of the 8° Brix sugar solution, were obtained in the test, showing the apparent superiority of this concentration over the others, particularly after the third week of incubation. When higher concentrations were compared with the 8° Brix sugar solution, i.e., 18° and 20° Brix, highly significant differences were obtained in favor of the 8° Brix.

Period of incubation

The period of incubation was for only three weeks, for it was not found profitable, particularly with molasses, to produce butyric acid beyond this period. The increase in the efficiency figures were only very slight in both the runs on molasses and sugar, especially with molasses. In molasses, a decrease in the efficiency figure was even observed after the third week in the 16° Brix solution, and in the 20° Brix, after the second week (table 3). In the sugar solutions, a decrease in the yields (efficiency) was observed after the third week, with the 18° and 20° Brix.

Observations

If the per cent yields of butyric acid based on total sugars are compared with those obtained by Arroyo (1934), the organism used in the present study was not as efficient as those isolated and employed by him. The kind of molasses sample used may also have influenced the yields.

SUMMARY

1. Production of butyric acid from molasses and sugar solutions was studied with a view to determine the right concentration of these agricultural products, and by-products for efficient production under local conditions.

2. Fifteen grams calcium carbonate per 100 grams molasses solution (11 per cent) and 4 grams calcium carbonate per 100 grams sugar solution (10 per cent) gave the best results.

3. Molasses culture solutions gave higher yield of butyric acid than sugar solutions. A 12° Brix molasses solution was the best. For sugar solutions, 8° Brix was the best, particularly in efficiency of butyric acid production.

4. The maximum period of incubation employed was three weeks, as it was found unprofitable to extend the time beyond this period.

5. The results in these experiments, though carried on only for short periods, indicate the possibility of producing butyric acid from raw sugar and final molasses.

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TABLE 1

Influence of calcium carbonate on the production of butyric acid from molasses and from sugar solutions [a]

a. On an 11-per cent molasses solution

CALCIUM CARBONATE PER 100 GRAM SAMPLE	SUGARS [b] CONSUMED	RESIDUAL SUGARS [b]	BUTYRIC ACID PRODUCED, PER 10 ML. SOLUTION	BUTYRIC ACID BASED ON SUGARS CONSUMED	BUTYRIC ACID [c] BASED ON TOTAL INITIAL SUGARS
grams	*grams*	*grams*	*grams*	*per cent*	*per cent*
Control	0.48	0.30	0.04	8.44 ± 0.26	5.17 ± 0.15
10	0.49	0.29	0.09	17.75 ± 1.06	11.20 ± 0.63
15	0.63	0.15	0.14	21.78 ± 0.53	17.49 ± 0.47
20	0.55	0.23	0.05	8.52 ± 0.09	6.26 ± 0.18
25	0.54	0.25	0.04	7.34 ± 0.31	5.08 ± 0.18

b. On a 10 per cent sugar solution

Control	0.26	0.62	0.04	16.14 ± 0.57	4.49 ± 0.11
4	0.27	0.61	0.08	31.08 ± 0.51	9.58 ± 0.16
8	0.25	0.64	0.04	17.39 ± 0.40	4.85 ± 0.05
12	0.27	0.61	0.04	14.63 ± 0.38	4.46 ± 0.11
16	0.21	0.67	0.05	22.37 ± 1.05	5.39 ± 0.23

[a] The time of incubation was two weeks.

[b] Sugars, expressed as invert sugar.

[c] The total initial sugars used in these experiments, per 10 ml. of solution, were: 0.7831 gm. for molasses, and 0.8817 gm. for sugar solution.

TABLE 2

Comparative influences of varying amounts of calcium carbonate on the production of butyric acid from molasses and from sugar solutions

a. On an 11-per cent molasses solution with the 15-gram treament as standard

AMOUNTS OF CALCIUM CARBONATE COMPARED	BUTYRIC ACID BASED ON INVERT SUGARS CONSUMED	BUTYRIC ACID BASED ON TOTAL ORIGINAL INVERT SUGARS
15 vs. control	21.78 ± 0.53 — 8.44 ± 0.26	17.49 ± 0.47 — 5.17 ± 0.15
Difference	13.34 ± 0.59 (HS) [a]	12.32 ± 0.49 (HS)
15 vs 10	21.78 ± 0.53 — 17.75 ± 1.06	17.49 ±0.47 — 11.20 ± 0.63
Difference	4.03 ± 1.19 (S) [b]	6.29 ± 0.72 (HS)
15 vs 20	21.78 ± 0.53 — 8.52 ± 0.09	17.49 ± 0.47 — 6.26 ± 0.18
Difference	13.26 ± 0.54 (HS)	11.23 ± 0.50 (HS)
15 vs 25	21.78 ± 0.53 — 7.34 ± 0.31	17.49 ± 0.47 — 5.08 ± 0.18
Difference	14.44 ± 0.61 (HS)	12.41 ± 0.50 (HS)

b. On a 10-per cent sugar solution with the 4-gram treatment as standard

4 vs control	31.08 ± 0.51 — 16.14 ± 0.57	9.58 ± 0.16 — 4.49 ± 0.11
Difference	14.24 ± 0.76 (HS)	5.09 ± 0.19 (HS)
4 vs 8	31.08 ± 0.51 — 17.39 ± 0.40	9.58 ± 0.16 — 4.85 ± 0.05
Difference	13.69 ± 0.63 (HS)	4.73 ± 0.17 (HS)
4 vs 12	31.08 ± 0.51 — 14.63 ± 0.38	9.58 ± 0.16 — 4.46 ± 0.11
Difference	16.45 ± 0.64 (HS)	5.12 ± 0.19 (HS)
4 vs 16	31.08 ± 0.51 — 22.37 ± 1.05	9.58 ± 0.16 — 5.38 ± 0.23
Difference	8.71 ± 1.17 (HS)	4.19 ± 0.25 (HS)

[a] HS—Highly significant
[b] S—Significant

TABLE 3

Effect of sugar concentration on the production of butyric acid from molasses and from sugar solutions

a. Molasses solution with 15 grams calcium carbonate

°BRIX	BUTYRIC ACID BASED ON SUGARS CONSUMED [a] AFTER			BUTYRIC ACID BASED ON TOTAL INITIAL SUGARS [b] AFTER		
	1st week	2nd week	3rd week	1st week	2nd week	3rd week
	per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
8	21.47 ± 0.37	17.94 ± 0.65	20.34 ± 0.68	8.51 ± 0.06	9.79 ± 0.55	14.93 ± 0.40
10	28.49 ± 0.70	20.51 ± 0.16	17.62 ± 1.16	10.84 ± 0.07	12.13 ± 0.10	13.05 ± 0.84
12	35.86 ± 1.08	23.85 ± 0.60	20.31 ± 0.37	12.81 ± 0.20	13.78 ± 0.24	14.95 ± 0.26
14	25.37 ± 0.23	14.82 ± 0.23	14.22 ± 0.48	7.74 ± 0.14	8.20 ± 0.16	9.81 ± 0.33
16	20.44 ± 0.28	13.65 ± 0.26	9.34 ± 0.24	5.55 ± 0.04	6.29 ± 0.11	5.98 ± 0.37
18	13.96 ± 0.11	12.34 ± 0.51	8.28 ± 0.51	5.59 ± 0.04	4.80 ± 0.14	4.96 ± 0.27
20	10.48 ± 0.07	6.82 ± 0.59	6.97 ± 0.61	2.66 ± 0.16	2.36 ± 0.14	3.07 ± 0.22

b. Sugar solutions with 4 grams calcium carbonate

8	18.93 ± 0.30	19.49 ± 0.29	19.47 ± 1.05	6.26 ± 0.05	11.37 ± 0.07	14.16 ± 0.67
10	27.50 ± 0.63	22.74 ± 0.28	16.95 ± 0.67	6.43 ± 0.03	9.55 ± 0.03	10.41 ± 0.48
12	36.67 ± 2.07	26.94 ± 0.34	20.52 ± 0.53	6.47 ± 0.03	8.86 ± 0.07	11.05 ± 0.30
14	24.61 ± 0.36	21.65 ± 0.18	15.81 ± 0.25	6.03 ± 0.05	8.87 ± 0.03	9.99 ± 0.20
16	28.05 ± 0.89	22.94 ± 0.46	16.71 ± 0.23	5.77 ± 0.03	8.39 ± 0.02	9.60 ± 0.12
18	31.83 ± 0.44	29.21 ± 0.06	18.11 ± 0.66	4.53 ± 0.01	6.88 ± 0.02	6.49 ± 0.16
20	26.68 ± 0.52	23.97 ± 0.42	16.45 ± 0.71	3.76 ± 0.02	6.32 ± 0.02	5.15 ± 0.30

[a] Sugars, as calculated, are expressed in terms of invert sugar.

[b] The total initial sugars of the different molasses and sugar solutions per 10 ml. of solution were:

8° Brix	0.57 gram	0.72 gram
10° "	0.73 "	0.96 "
12° "	0.85 "	1.03 "
14° "	0.97 "	1.26 "
16° "	1.15 "	1.45 "
18° "	1.31 "	1.59 "
20° "	1.44 "	1.74 "

TABLE 4

Comparative summary of the influences of varying molasses and sugar concentrations on the production of butyric acid

a. Molasses solutions with the 12° Brix as the standard

CONCENTRATIONS (°BRIX) COMPARED	BUTYRIC ACID BASED ON SUGARS CONSUMED AFTER		
	First week	Second week	Third week
	per cent	*per cent*	*per cent*
12 vs 8	35.86 ± 1.08 — 21.47 ± 0.37	23.85 ± 0.60 — 17.94 ± 0.65	20.31 ± 0.37 — 20.34 ± 0.47
Difference	14.39 ± 1.14 (HS) [a]	5.91 ± 0.88 (HS)	—0.03 ± 0.60 (I) [c]
12 vs 10	35.86 ± 1.08 — 28.49 ± 0.70	23.85 ± 0.60 — 20.51 ± 0.16	20.31 ± 0.37 — 17.62 ± 1.16
Difference	7.37 ± 1.29 (HS)	3.34 ± 0.62 (S) [b]	2.69 ± 1.22 (I)
12 vs 14	35.86 ± 1.08 — 25.37 ± 0.23	23.85 ± 0.60 — 14.82 ± 0.23	20.31 ± 0.37 — 14.22 ± 0.68
Difference	10.49 ± 1.10 (HS)	9.03 ± 0.64 (HS)	6.09 ± 0.61 (HS)
12 vs 16	35.86 ± 1.08 — 20.44 ± 0.28	23.85 ± 0.60 — 13.65 ± 0.26	20.31 ± 0.37 — 9.34 ± 0.24
Difference	15.42 ± 0.11 (HS)	10.20 ± 0.65 (HS)	10.97 ± 0.44 (HS)
12 vs 18	35.86 ± 1.08 — 13.96 ± 0.11	23.85 ± 0.60 — 12.34 ± 0.51	20.31 ± 0.37 — 8.28 ± 0.51
Difference	21.90 ± 1.09 (HS)	11.51 ± 0.79 (HS)	12.03 ± 0.63 (HS)
12 vs 20	35.86 ± 1.08 — 10.48 ± 0.07	23.85 ± 0.60 — 6.82 ± 0.59	20.31 ± 0.37 — 6.97 ± 0.61
Difference	25.38 ± 1.08 (HS)	17.03 ± 0.84 (HS)	13.34 ± 0.71 (HS)

b. Sugar solutions with the 8° Brix as the standard

8 vs 10	18.93 ± 0.30 — 27.50 ± 0.63	19.49 ± 0.29 — 22.74 ± 0.28	19.47 ± 1.05 — 16.95 ± 0.67
Difference	—8.57 ± 0.70 (HS) [a]	— 3.25 ± 0.40 (HS)	2.52 ± 1.25 (I) [c]
8 vs 12	18.93 ± 0.30 — 36.67 ± 2.07	19.49 ± 0.29 — 26.94 ± 0.34	19.47 ± 1.05 — 20.52 ± 0.53
Difference	— 17.74 ± 2.09 (HS)	— 7.45 ± 0.45 (HS)	— 1.05 ± 1.18 (I)
8 vs 14	18.93 ± 0.30 — 24.61 ± 0.36	19.49 ± 0.29 — 21.65 ± 0.18	19.47 ± 1.05 — 15.81 ± 0.25
Difference	— 5.68 ± 0.47 (HS)	—2.16 ± 0.34 (S) [b]	3.66 ± 1.08 (SS) [d]
8 vs 16	18.93 ± 0.30 — 28.05 ± 0.89	19.46 ± 0.29 — 22.94 ± 0.46	19.47 ± 1.05 — 16.71 ± 0.23
Difference	— 9.12 ± 0.94 (HS)	— 3.45 ± 0.54 (S)	2.76 ± 1.07 (I)
8 vs 18	18.93 ± 0.30 — 31.83 ± 0.44	19.49 ± 0.29 — 29.21 ± 0.06	19.47 ± 1.05 — 18.11 ± 0.66
Difference	— 12.90 ± 0.53 (HS)	— 9.72 ± 0.30 (HS)	1.36 ± 1.24 (I)
8 vs 20	18.93 ± 0.30 — 26.68 ± 0.52	19.49 ± 0.29 — 23.97 ± 0.42	19.47 ± 1.05 — 16.45 ± 0.71
Difference	— 7.75 ± 0.60 (HS)	— 4.48 ± 0.51 (HS)	3.02 ± 1.27 (I)

[a] HS—Highly significant
[b] S—Significant
[c] I—Insignificant
[d] SS—Slightly significant

TABLE 4 (Continued)

CONCENTRATIONS (°BRIX) COMPARED	BUTYRIC ACID BASED ON TOTAL ORIGINAL SUGARS AFTER		
	First week	Second week	Third week
	per cent	*per cent*	*per cent*
12 vs 8	12.81 ± 0.20 — 8.51 ± 0.06	13.78 ± 0.24 — 9.79 ± 0.55	14.95 ± 0.26 — 14.95 ± 0.40
Difference	4.30 ± 0.21 (HS)	3.99 ± 0.60 (S)	0.00 ± 0.48 (I)
12 vs 10	12.81 ± 0.20 — 10.84 ± 0.07	13.78 ± 0.24 — 12.13 ± 0.10	14.95 ± 0.26 — 13.05 ± 0.84
Difference	1.97 ± 0.22 (HS)	1.65 ± 0.24 (S)	1.90 ± 0.88 (I)
12 vs 14	12.81 ± 0.20 — 7.74 ± 0.14	13.78 ± 0.24 — 8.20 ± 0.16	14.95 ± 0.26 — 9.81 ± 0.33
Difference	5.07 ± 0.24 (HS)	5.58 ± 0.29 (HS)	5.14 ± 0.42 (HS)
12 vs 16	12.81 ± 0.20 — 5.55 ± 0.04	13.78 ± 0.24 — 6.29 ± 0.11	14.95 ± 0.26 — 5.98 ± 0.37
Difference	7.26 ± 0.20 (HS)	7.49 ± 0.26 (HS)	8.97 ± 0.45 (HS)
12 vs 18	12.81 ± 0.20 — 3.59 ± 0.04	13.78 ± 0.24 — 4.80 ± 0.14	14.95 ± 0.26 — 4.96 ± 0.27
Difference	9.22 ± 0.20 (HS)	8.98 ± 0.28 (HS)	9.99 ± 0.37 (HS)
12 vs 20	12.81 ± 0.20 — 2.66 ± 0.16	13.78 ± 0.24 — 2.36 ± 0.14	14.95 ± 0.26 — 3.07 ± 0.22
Difference	10.15 ± 0.26 (HS)	11.42 ± 0.28 (HS)	11.88 ± 0.34 (HS)
8 vs 10	6.26 ± 0.05 — 6.43 ± 0.03	11.37 ± 0.07 — 9.55 ± 0.03	14.16 ± 0.67 — 10.41 ± 0.48
Difference	— 0.17 ± 0.06 (I)	1.82 ± 0.08 (HS)	3.75 ± 0.82 (S)
8 vs 12	6.26 ± 0.05 — 6.47 ± 0.03	11.37 ± 0.07 — 8.86 ± 0.07	14.16 ± 0.67 — 11.05 ± 0.30
Difference	— 0.21 ± 0.06 (SS)	2.51 ± 0.99 (I)	3.11 ± 0.73 (S)
8 vs 14	6.26 ± 0.05 — 6.03 ± 0.05	11.37 ± 0.07 — 8.87 ± 0.03	14.16 ± 0.67 — 9.99 ± 0.20
Difference	0.23 ± 0.07 (HS)	2.50 ± 0.08 (HS)	4.17 ± 0.70 (S)
8 vs 16	6.26 ± 0.05 — 5.77 ± 0.03	11.37 ± 0.07 — 8.39 ± 0.02	14.16 ± 0.67 — 9.60 ± 0.12
Difference	0.49 ± 0.06 (HS)	2.98 ± 0.07 (HS)	4.56 ± 0.68 (S)
8 vs 18	6.26 ± 0.05 — 4.53 ± 0.01	11.37 ± 0.07 — 6.88 ± 0.02	14.16 ± 0.67 — 6.49 ± 0.16
Difference	1.73 ± 0.05 (HS)	4.49 ± 0.07 (HS)	7.67 ± 0.69 (HS)
8 vs 20	6.26 ± 0.05 — 3.76 ± 0.02	11.37 ± 0.07 — 6.32 ± 0.02	14.16 ± 0.67 — 5.15 ± 0.30
Difference	2.50 ± 0.05 (HS)	5.05 ± 0.07 (HS)	9.01 ± 0.73 (HS)

EXPERIMENTAL TRANSMISSION OF THE MOSAIC OF CANNA INDICA [1]

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WITH TWO PLATES

The mosaic of *Canna indica* Linn. was probably first reported by Fukushi (1932) in 1928 in Japan. Although the disease has perhaps existed in Davao, Mindanao, Philippines, for many years, *Canna indica* mosaic was reported only in 1937 (Ocfemia, 1937). Mosaicked *Canna indica* plants were widespread along the road and among abacá plants in plantations in Bankas on the way to Tunkalan, Davao. Because of the similarity of the symptoms of the *Canna* mosaic to those of abacá, specimens were brought to Los Baños for study.

THE DISEASE

Importance

The mosaic of *Canna indica* in abacá plantations in Davao is of importance because it may be transmitted to abacá, ornamental varieties of *Canna* species and *Canna edulis* Ker. Ocfemia and Celino (1938) showed that abacá mosaic is transmitted by *Aphis gossypii* Glover and two other species of aphids to *Canna indica*. It is important to know if *Canna indica* mosaic is transmissible to abacá and what agents are responsible for its transmission.

Symptoms

On leaves: early stage of infection. The earliest symptom of mosaic of *Canna indica* may be seen in a two-week old aphid-transmission experiment. This symptom consists of short fine chlorotic lines connecting any two adjacent veins from the midrib to the margin of the youngest unfurled leaf. Any adjacent veins may be bridged by one or several of these chlorotic lines which are irregularly distanced

[1] Experiment Station contribution No. 1441. Read before the Los Baños Biological Club, June 26, 1941.
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from one another. With the veins thus connected by cross-chlorotic dashes, the symptom takes the form of a network of discoloration. As the first infected leaf grows older, the chlorotic cross dashes become broader until they are about one millimeter. Some of the chlorotic lines may develop into more conspicuous spindle-shaped areas, with their long axis parallel with the veins. In some parts of the leaf, the chlorotic areas which flank neighboring veinlets coalesce.

Advanced stage of infection. On leaves that appear in later stages of the disease, the chlorotic areas are broad and long. They appear either as regular or irregular stripes parallel with the veins and extend from the midrib to the margin. The stripes may not be solidly chlorotic because they may include patches of green. Owing perhaps to retarded development of the chlorotic areas, the infected leaf may become wrinkled or curled (pl. 1, fig. 1). Chlorotic dot-like or spindle-shaped areas may also be irregularly distributed on the green portions of the leaf blades. In still more advanced cases of infection the yellowed areas become necrotic and turn rusty brown. The necrotic effect and the rusty-brown color of the streaks are the common symptoms of *Canna indica* mosaic on the cultivated ornamental varieties of *Canna.*

On the stems and leaf sheaths. The stems and leaf sheaths of infected *Canna indica* appear more lightly colored than those of healthy plants. Irregular green patches are formed on the chlorotic background of the stem. As a consequence of the premature drying of leaves, the lower parts of the stems of diseased *Canna indica* appear to be more slender than those of healthy plants.

On flowers. The sepals of the flowers of mosaicked *Canna indica* are more lightly colored than those of the healthy plants. The petals are also generally more lightly colored and they have pale red longitudinal bands running from the base to the tip of the petals. In some cases they form almost uniform streaks about one millimeter in width. Dark red, oblong or more or less elongate areas may be seen on the pale red or orange colored parts. The yellow streaks on the stamens of mosaicked *Canna indica* are larger and more conspicuous than those on the flowers of healthy plants. The pistil may be almost orange throughout.

In the flowers of the cultivated varieties of *Canna* species, broken and irregular-shaped orange streaks are present. These streaks arise from the base of the petals and disappear as they approach the margins of the petals. The orange streaks produce a beautiful effect

on the red petals of the cultivated ornamental *Canna* species. The streaks, however, are not sufficiently conspicuous to attract attention.

On fruits. The fruit of *Canna indica* infected either with the *Canna* mosaic or abacá mosaic also shows irregular mottling of pale yellow and green. The mottling, however, is not as distinct as that on the leaves.

Symptoms produced by the Canna indica-mosaic virus on abacá seedlings. On abacá seedlings to which *Canna indica* mosaic has been transmitted by *Aphis gossypii,* the symptom consists of greenish yellow streaks with more or less indefinite borders (pl. 2, fig. 1). The streaks begin from the midrib of the leaf and end at the margin. At first the streaks are hardly a millimeter in width with their borders diffusing with the normal green color. Lateral coalescence of several narrow streaks results in the production of yellowish orange areas varying from two to ten and even fifteen millimeters in width by ten to twenty millimeters in length.

The symptoms of the advanced stage of *Canna indica* mosaic on *Canna indica* seedlings (pl. 1, fig. 1) and those of the early and advanced stages of infection of abacá seedlings are different from those shown by abacá mosaic on *Canna indica* (pl. 1, fig. 2). The young symptoms shown by abacá mosaic on *Canna indica* and on abacá seedlings are described by Celino (1940).

Symptoms produced by the abacá-mosaic virus on Canna indica. In aphid-transmission experiments of abacá mosaic to *Canna indica,* the first symptom may be noted on the first leaf that unfurls, ten to fifteen days after introduction of infective aphids to the experimental plant (pl. 1, fig. 2 and pl. 2, fig. 4). A number of dot-like yellowish areas ranging in size from one-half to one millimeter in diameter may be seen. About twenty days after the appearance of numerous yellowish spots, spindle-shaped yellowed streaks are formed. The larger spots vary from one and one-half millimeters to two and one-half by three to fifteen or more millimeters. Some of the streaks may extend from the midrib to the margin of the leaves.

TRANSMISSION EXPERIMENTS AND RESULTS

Through seeds

Experiment 1. On January 4, 1939, seventeen seeds gathered from a mosaicked *Canna indica* were planted in a pot of sterile soil. None of the sixteen seeds that germinated after 41 to 55 days had symptoms of mosaic.

Experiment 2. On January 19, 1939, twenty-three seeds gathered from healthy *Canna indica* plants and thirty-three seeds from mosaicked plants were planted in two pots of sterile soil. Of the seeds that came from healthy plants none of the twenty-two that emerged showed symptoms of mosaic. Similarly, all of the twenty seeds from mosaicked plants that germinated were free from the disease.

Experiment 3. On January 22, 1941, sixty seeds from *Canna indica* infected with abacá mosaic and one hundred and twelve seeds from *Canna indica* with *Canna* mosaic were planted in two large pots of sterile soil. Only a few of the seeds of both lots were sound and healthy looking. In order to see whether discolored, wrinkled and poor-looking seeds carried the disease, all of the seeds were planted. The seedlings began to emerge on January 30, 1941. On February 26, 1941, only seven of the sixty seeds gathered from *Canna* plants with the abacá mosaic emerged, and only six of the one hundred and twelve seeds from *Canna indica* infected with the *Canna* mosaic. All of the seedlings that emerged were free from the mosaic symptoms.

The results of the germination experiments indicate that seeds of *Canna indica* gathered from plants infected either with *Canna* mosaic or abacá mosaic do not carry the virus.

Transmission by insects

In all insect-transmission experiments, the method described by Ocfemia in 1930 for transferring *Pentalonia nigronervosa* Coq. from the source of inoculum to the experimental abacá seedlings was followed. The aphids used were: *Aphis gossypii* Glover [3], *Pentalonia nigronervosa, Aphis laburni* Kaltenbach, *Aphis maidis* Fitch. and *Rhopalosiphum nympheae* (Linné).

Experimental transmission of Canna indica mosaic to abacá with the use of Aphis gossypii Glover as vector

Experiment 1. On September 20, 1940, adult *Aphis gossypii* Glover from healthy cotton plants were allowed to feed on a mosaicked *Canna indica* for about two hours. Thirty of these aphids were transferred to an abacá seedling which was two and one-half months old. The seedling with the aphids on it was placed in an insect-proof chamber to protect the insects from predators. Another healthy

[3] The aphids were determined in the Department of Entomology, College of Agriculture.

abacá seedling on which no aphids were introduced was used as control.

On October 5, 1940, after a period of incubation of fifteen days, the first symptom of mosaic was noticed on the experimental abacá seedling whereas the check remained healthy.

Experiment 2. On October 7, 1940, twenty-five *Aphis gossypii* were placed on each of nine healthy three-month old abacá seedlings. The same number of healthy seedlings, without aphids, served as control.

All abacá seedlings on which infective *Aphis gossypii* were allowed to feed were infected after eleven to twenty-six days of incubation. None of the checks developed the disease.

In addition to the fact that *Aphis gossypii* readily transmitted *Canna indica* mosaic to abacá seedlings, this aphid also recovered the virus from the infected abacá seedlings and transmitted it to *Canna indica.*

The shortest time of feeding of Aphis gossypii on mosaicked Canna indica necessary to acquire the virus for transmission to abacá seedlings

Experiment 1. On October 19, 1940, each of six healthy abacá seedlings, three months old, was used for colonizing twenty adult *Aphis gossypii* which had fed for forty-five minutes on mosaicked *Canna indica.* All plants became diseased after incubation periods ranging from eight to fourteen days.

Experiment 2. On October 21, 1940, twenty aphids that had fed on mosaicked *Canna indica* for thirty minutes were transferred to each of three healthy abacá seedlings. All abacá seedlings developed mosaic after periods of incubation from seven to twelve days.

Experiment 3. On October 22, 1940, from twenty to twenty-five *Aphis gossypii* which had fed on diseased *Canna indica* for twenty minutes were transferred to each of two healthy abacá seedlings. Eleven days later both seedlings were infected with mosaic. Experiment 3 was repeated on November 1 and on November 5, 1940. In each repetition two plants were used. After thirteen days of incubation the experimental seedlings developed mosaic.

Experiment 4. On October 28, 1940, after *Aphis gossypii* had been allowed to feed for ten minutes on diseased *Canna indica,*

twenty to twenty-five of them were transferred to each of two healthy abacá seedlings. On November 19, 1940, three seedlings were used; on November 25, two seedlings; and on November 26, 1940, two seedlings. Infection appeared after eighteen to twenty-nine days of incubation.

Experiment 5. After feeding for five minutes on diseased *Canna indica,* twenty to twenty-five *Aphis gossypii* were transferred to each of five abacá seedlings on December 2, and five seedlings on December 28, 1940. Four of those used on December 2 were infected after nineteen to twenty-five days of incubation. Two of the abacá seedlings used on December 28, 1940, developed mosaic symptoms sixteen to twenty-five days later.

In these five experiments it was noted that it took sometime before *Aphis gossypii* settled down to feed. The writers believe that *Aphis gossypii* is capable of obtaining the virus of *Canna indica* mosaic in less than five minutes of feeding and of transmitting it to abacá seedlings.

Doolittle and Walker (1928) found that *A. gossypii* can transmit cucumber mosaic in five minutes of feeding on diseased plant. Stahl and Carsner (1923) report that *Euttetix tenellus* Baker can obtain the virus of curly leaf of sugar beet in ten to twenty minutes. Celino (1940) found that it takes *Rhopalosiphum nympheae* two hours to become infective with the virus of abacá mosaic.

As *Aphis gossypii* readily transmits *Canna indica* mosaic to *Canna indica* and abacá seedlings, it may be an important vector in the field.

The smallest number of Aphis gossypii necessary to transmit Canna indica mosaic to abacá

Experiment 1. Fifteen adult *Aphis gossypii* that had fed on diseased *Canna indica* for about an hour were transferred to each of three healthy abacá seedlings on October 24, to one seedling on October 25, and to each of three young abacá plants on November 4, 1940. All seven seedlings were infected after periods of incubation ranging from ten to twenty-four days.

Experiment 2. On October 24, 1940, ten virus-laden aphids were transferred to each of three healthy abacá seedlings which were three months old. After a period of incubation from fourteen to thirty-three days, all of the three seedlings were infected.

On October 30, 1940, ten infective aphids were placed on each of two healthy abacá seedlings. One of the plants was infected after eleven days of incubation and the other after twelve days.

Experiment 3. Five infective aphids were placed on each of two healthy three-month old abacá seedlings on October 24, on two seedlings on October 28, on nine seedlings on November 6, on two on November 19, on two on November 25, and on two on November 26, 1940. Nine to thirty-five days later all of the experimental abacá seedlings were mosaicked.

Ocfemia (1930) found that five adult *Pentalonia nigronervosa* can transmit bunchy-top of abacá. The writers noted that five adult *Aphis gossypii* can also obtain sufficient virus of *Canna indica* mosaic to transmit it to abacá seedlings. Celino (1940) reports that eight adult *Rhopalosiphum nympheae* could transmit sufficient abacá-mosaic virus to cause infection of abacá seedlings.

Retention of the virus by Aphis gossypii

Experiment 1. On December 13, 1940, forty *Aphis gossypii* which had fed on a mosaicked *Canna indica* for about an hour, were transferred to each of three sets of two abacá seedlings each. After the aphids were allowed to feed on the first two seedlings for about two hours they were transferred to the second set of two seedlings. After two hours on the second seedlings the aphids were transferred to the third set of two seedlings.

Seventeen days later, the first set of two seedlings showed mosaic, whereas the second and the third sets remained healthy.

Experiment 2. On January 13, 1941, the experiment was repeated in duplicates and after an incubation period of nineteen to twenty-one days, the seedlings of the first set showed mosaic. The seedlings in the second and third sets remained healthy.

Experiment 3. On February 4, 1941, another three sets of two seedlings each were used. Thirteen days later the seedlings in the first set became diseased whereas the seedlings in the second and third remained healthy.

From the results of these three experiments it is concluded that *Aphis gossypii* loses all of the virus of *Canna indica* mosaic in the first feeding on healthy abacá seedlings.

Failure of Aphis gossypii to infect abacá seedlings with Canna indica mosaic if not allowed to feed on the host immediately

Experiment 1. On December 6, 1940, thirty infective aphids were kept for one hour in a sterile test tube and then transferred to an abacá seedling. Five other seedlings were similarly treated. To a check seedling, thirty infective aphids were transferred directly from the diseased *Canna indica.*

On January 8, 1941, the control became mosaicked, whereas the six experimental seedlings remained healthy.

Experiment 2. On January 21, 1941, thirty infective aphids were kept in a sterile test tube for thirty minutes and then placed on a young abacá seedling. Five other seedlings were similarly treated. The experiment was repeated on February 11, 1941, with the use of four abacá seedlings.

On February 9, 1941, one of the seedlings used on January 21 showed mosaic but the rest did not.

The results of these two experiments seem to show that *Aphis gossypii* cannot transmit *Canna indica* mosaic to abacá if it is not allowed to feed within thirty minutes. The *Canna indica* mosaic virus differs from the virus of bunchy-top of abacá (Ocfemia and Buhay, 1934) in that it does not require a period of incubation in the body of the vector. In the control of *Canna indica* mosaic, this behavior of the virus is of importance because it does away with the need for destroying the insect vectors except when these are on or near the susceptible plants.

Transmission of Canna indica mosaic to abacá seedlings by Aphis maidis Fitch

On October 31, 1940, thirty adult *Aphis maidis* Fitch. which had fed for one hour on mosaicked *Canna indica* were transferred to each of two healthy abacá seedlings. Eight other plants were similarly treated on December 21, 1940. On January 8, 1941, six additional abacá seedlings were used for colonizing the same number of infective *Aphis maidis.*

On November 26, 1940, one of the plants used on October 31, 1940, developed mosaic. On January 8, 1941, one of those used on December 21, 1940, became mosaicked; another one was infected on February 2, 1941, and the third plant was diseased on February 9, 1941. Of those used on January 8, 1941, one was infected on January 20, 1941.

The result of this experiment indicates that *Aphis maidis* can transmit the virus of *Canna indica* mosaic to abacá seedlings. Celino and Ocfemia (1941) transmitted abacá mosaic with *Aphis maidis* not only to abacá but also to corn. Furthermore, they found that *Aphis maidis* could recover the virus from corn and transmit it back to abacá seedlings and corn seedlings.

Transmission of Canna indica mosaic to ornamental varieties of Canna species

On November 23, 1940, twenty-five infective *Aphis gossypii* were transferred to a cultivated ornamental *Canna* species *D*[4]; on November 29, to each of two different varieties of ornamental *Canna, F* and *H;* and on December 26, to another ornamental *Canna, I.*

On December 24, 1940, cultivated ornamental *Canna* species *H* showed infection after an incubation period of twenty-five days. On January 17, 1941, ornamental *Canna I* showed mosaic after an incubation period of twenty-two days. Celino (1940) could not transmit abacá mosaic to ornamental *Canna* species with *Rhopalosiphum nympheae.*

Transmission to Canna edulis Ker.

On December 12, 1940, three plants of *Canna edulis* Ker. were used for colonizing *Aphis gossypii* taken from diseased *Canna indica.* One *Canna edulis* was used in a transmission experiment on December 13, and two plants on December 28, 1940.

One of the plants used on December 12, 1940, showed mosaic on January 9, 1941, and one of those used on December 28, 1940, was infected on January 17, 1941. This result indicates that *Canna indica* mosaic may be transmitted to *Canna edulis.* Celino (1940) failed to transmit abacá mosaic to *Canna edulis* with *Rhopalosiphum nympheae* as vector.

Failure of transmission to cucumber

On November 5, 1940, a one-week-old cucumber seedling was used for colonizing twenty-five *Aphis gossypii* which had fed for one to two hours on diseased *Canna indica.* On November 25, 1940, four four week-old cucumber seedlings and on November 29, 1940, two other seedlings were used for colonizing the same number of infec-

[4] Letters *D, F, H* and *I* are used for designating the unidentified varieties of ornamental *Canna* used. The plants were obtained from the Department of Agronomy.

tive *Aphis gossypii.* Even after months of incubation, however, none of the cucumber plants showed mosaic infection.

Failure of transmission to cotton

On December 26, 1940, two seedlings of Pima Egyptian and two seedlings of King 2266 were used in a transmission experiment with *Canna indica* mosaic by *Aphis gossypii.* The experiment was repeated on January 23, 1941, with the use of three seedlings of each of the two varieties of cotton. Even after a prolonged incubation period, however, no transmission of the disease to cotton was noted. Celino (1940) also failed to transmit abacá mosaic with *Aphis gossypii* to cotton seedlings.

Failure of transmission of Canna indica mosaic by Rhopalosiphum nympheae, Pentalonia nigronervosa, and Aphis laburni

Ocfemia and Celino (1938) and Celino (1940) transmitted abacá mosaic with *Aphis gossypii* and *Rhopalosiphum nympheae* as vectors. In addition to these aphids, Celino and Ocfemia (1941) transmitted abacá mosaic with the use of *Aphis maidis* and another species of *Rhopalosiphum* probably *R. prunifoliae* Fitch. These authors, however, failed to communicate abacá mosaic with *Aphis laburni* and *Pentalonia nigronervosa.*

In experiments to transmit *Canna indica* mosaic with the use of other aphids, the writers also tried *Rhopalosiphum nympheae, Pentalonia nigronervosa,* and *Aphis laburni.* The results of the experiments, however, showed that *Canna indica* mosaic cannot be transmitted by these three aphids.

Failure of transmission of Canna indica mosaic to abacá through the uninjured roots

On November 18, 1940, the roots of fifteen young abacá seedlings, variety Ilayas were placed in test tubes containing sap extract of *Canna indica* mosaic virus diluted with an equal volume of sterile water. Two other abacá seedlings were similarly treated with equal parts of Crone's solution and water-diluted extract of mosaicked *Canna indica.* Seven of the fifteen plants with their roots immersed in the diluted sap extract were planted in pots of soil after immersion for twenty-four hours, and the other eight seedlings after forty-eight hours. The seedlings in the water-diluted sap and Crone's solution were planted in pots of soil also after forty-eight hours.

The plants were observed for appearance of mosaic but none of the seedlings showed infection even after several weeks.

Failure of transmission of abacá mosaic with the use of powdered glass abrasive

Before attempting to transmit *Canna indica* mosaic by mechanical methods, tests were made on abacá with abacá mosaic because abacá has more sap than *Canna indica*.

Experiment 1. On November 18, 1940, the upper surface of the youngest expanded leaf of each of four abacá seedlings was wiped off with a wad of cotton wet with undiluted sap of mosaicked abacá. Before being used, the cotton was pressed against fine glass powder to produce abrasions on the leaf. The plants were set aside for observation but no infection took place even after several weeks.

Experiment 2. On December 20, 1940, the juice was diluted with one-tenth molar solution of monobasic potassium phosphate and then rubbed on the upper surface of the leaf of two abacá seedlings, and on the lower surface of the leaf of two other abacá seedlings. None of the plants became infected either.

Inoculation of abacá mosaic and Canna indica mosaic viruses through the use of glass wool

Experiment 1. On December 20, 1940, eight and one-half grams of macerated leaves of mosaicked abacá were mashed. Four milliliters of M/10 monopotassium phosphate were added to the mash and then rubbed on the upper surface of the second youngest leaf of each of ten healthy abacá seedlings by means of a wad of cotton. After applying the juice extract, the leaf was rubbed with glass wool in order to injure the surface of the leaf and insure contact of the juice with the exposed tissues of the abacá leaf. Some of the pieces of glass wool were buried in the leaf. After the surface of the leaves were rubbed with the glass wool, the liquid from the macerated juice was squeezed over the injured surface. Distilled water was atomized over the inoculated leaves three times at intervals of about thirty minutes to prevent rapid drying of the sap.

In a duplicate test, juice of mosaicked *Canna indica* was used for inoculation. On January 15, 1941, one of the inoculated abacá plants was infected after an incubation period of twenty-six days.

Experiment 2. On January 7, 1941, experiment 1 was repeated and seven abacá seedlings were used.

Experiment 3. On January 9, 1941, five abacá seedlings were inoculated with the sap of mosaicked *Canna indica.* In this experiment instead of monopotassium phosphate, an equal volume of sterile water was added.

Experiment 4. On January 21, 1941, ten abacá seedlings were inoculated with juice extract from mosaicked *Canna indica.*

Inasmuch as only one of the abacá seedlings inoculated with *Canna indica* mosaic virus in experiment 1 became infected and none of those used in succeeding experiments became diseased, the result is regarded as inconclusive.

SUMMARY

1. The mosaic of *Canna indica* Linn. in Davao attracted attention in May, 1937. The disease resembles the mosaic of abacá.

2. *Canna indica* mosaic produces on the leaves irregular pale yellow stripes parallel with the veins. The stripes may extend from the midrib to the margin of the leaves. The leaves are more or less wrinkled and curled. The chlorotic areas often turn rusty-brown. Yellowish bands may be noted on the stems and on the sepals and petals of the flowers. Mottling of the fruit is usually not as distinct as that of the leaves.

3. The symptoms of the advanced stage of *Canna indica* mosaic on *Canna indica* seedlings and the symptoms of the early and advanced stages of *Canna indica* mosaic on abacá seedlings differ from those produced by abacá mosaic on *Canna indica* and on abacá.

4. *Canna indica* mosaic may be transmitted to abacá by *Aphis gossypii* Glover and *Aphis maidis* Fitch. *Aphis gossypii* can also transmit *Canna indica* mosaic to *Canna edulis* and two varieties of ornamental *Canna* species.

5. Although *Aphis gossypii* can transmit *Canna indica* mosaic to varieties of ornamental *Canna* species and *Canna edulis,* this aphid cannot transmit the mosaic of abacá to these plants. Neither can *Rhopalosiphum nymphaeae* (Linné) transmit abacá mosaic to ornamental *Canna* species and *Canna edulis.*

6. *Aphis gossypii* can transmit *Canna indica* mosaic to abacá seedlings after feeding for about five minutes on diseased *Canna indica.* Five *Aphis gossypii* are sufficient to effect transmission of *Canna indica* mosaic to abacá seedlings.

7. A virus-laden *Aphis gossypii* loses all of the virus when it feeds on the first plant. If *Aphis gossypii* that had fed on mosaicked

Canna indica are placed in a test tube for an hour they cannot infect abacá seedlings with the mosaic of *Canna indica.*

8. *Canna indica* mosaic cannot be transmitted by *Aphis laburni* Kaltenbach, *Pentalonia nigronervosa* Coq., and *Rhopalosiphum nympheae.*

9. Whether *Canna indica* mosaic is identical with the abacá mosaic or not, mosaicked *Canna indica* is a source of inoculum for infecting abacá with mosaic disease.

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DESCRIPTION OF PLATES

PLATE 1

Mosaic of *Canna indica* Linn. 1. A leaf infected with the *Canna indica* mosaic that occurs in Davao. 2. A leaf of *Canna indica* infected with the abacá mosaic transmitted by *Aphis gossypii* Glover. Note the larger mottled areas on the leaf in figure 1, the more conspicuous curling of its blade, and the more undulated surface of the leaf than that of the foliage shown in figure 2. Photograph by the Photographic Division, College of Agriculture.

PLATE 2

Different patterns produced by mosaic in the advanced stage of the disease: (1) *Canna indica* mosaic on an abacá leaf, and (2) abacá mosaic on abacá leaf. Both leaves were taken from abacá seedlings of the same variety and age. Figures 1 and 2 about one-half natural size. (3) *Canna indica* mosaic on *Canna indica* leaf, and (4) abacá mosaic on *Canna indica* leaf. Figures 3 and 4 about one-third natural size. All photographs by the Photographic Division, College of Agriculture.

Plate 1

Plate 2

HEMATOLOGICAL STUDIES ON CATTLE: IV. VARIATIONS OF HEMOGLOBIN AND WHITE BLOOD CELLS IN THE PHILIPPINE NATIVE BREED OF CATTLE[1]

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Of the Department of Animal Husbandry

WITH TWO TEXT FIGURES

Studies on the fluctuations in hemoglobin of the Indian Nellore breed of cattle carried out in the College of Agriculture during a period of one year showed that the hemoglobin indices follow bimodal curves. The first curve is of seven months duration, starting in October and ending in April, with the mode in January. The second curve started in May and ended in September, with the mode in July. The rise and fall in hemoglobin indices were gradual and regular. The absence of significant correlations in Nellore cattle between hemoglobin and relative humidity was interpreted to mean that animals which have become adapted to a given environment, such as the Nellore in the Philippines, are less affected by environmental factors obtaining therein. No correlation was found between hemoglobin and body temperature which indicated that normal fluctuation of body temperature does not markedly affect the hemoglobin indices. As it seems important to follow these studies on Philippine Native cattle, this investigation was conducted to determine (*a*) the hemoglobin indices in Philippine Native cattle throughout the entire year, (*b*) if the fluctuations follow a regular curve in the different months of the year, (*c*) whether these fluctuations are affected by relative humidity and atmospheric temperature, and (*d*) whether the fluctuations are correlated with body temperature and different kinds of white blood corpuscles.

MATERIALS AND METHODS

Six mature Philippine Native cattle, three castrated males and three females, were used in this experiment. The color of these animals was uniformly light red. The care and management of the

[1] Experiment Station contribution No. 1442. The greater part of the data in this paper was used in a thesis presented by the junior author for graduation in March, 1940, with the degree of Bachelor of Science in Agriculture. The thesis was prepared under the direction of the senior author.

animals were alike throughout the whole period of experimentation. A complete list of the animals used in this experiment is given below:

NAME AND HERD NUMBER OF ANIMALS	SEX	AGE AT COMMENCEMENT OF EXPERIMENT		WEIGHT AT THE BEGINNING OF THE EXPERIMENT
		Years	Months	
				kgm.
Magno, 280	Male	2	8	283.6
Sofronio, 285	Male	2	8	255.5
Silvestre, 284	Male	3	4	320.9
Maxima, 275	Female	3	0	190.9
Marta, 277	Female	2	11	185.5
Sampaguita, 283	Female	3	11	278.2

Technique used in sampling blood and in making differential counts

Hemoglobin. The technique used for the sampling of blood for hemoglobin determinations was the same as that reported in previous papers of this series; and as in previous work, the hemoglobin determinations were made at intervals of one week. The time of sampling was at two o'clock on Saturday afternoons.

Differential white blood cell counts. Blood smears were made from each animal at the time of sampling and were stained with Wright's stain within one hour and were examined with a two-milimeter oil immersion objective. The different varieties of white blood cells most commonly found were lymphocytes, large mononuclears, polymorphonuclears, eosinophiles, and mast cells. In order to obtain the relative proportion of varieties of leucocytes, at least 300 of these white blood cells were counted. The percentage of each variety was expressed in relation to the total number in the film.

Relative humidity and atmospheric temperature. Readings on the relative humidity and the temperature of the air during the operation were obtained from the meteorological records for the College of Agriculture Campus at the Department of Agricultural Botany.

Condition of the animals and their body temperatures. The live weight of the animals was taken on the last day of each month throughout the duration of the experiment, and the general health and condition of the animals were observed every week. The body

temperatures were taken with an "Empire" thermometer held in the rectum, in no case less than three minutes.

RESULTS OF THE EXPERIMENT

Monthly weights of animals

The monthly weights of the animals used varied during the period of the experiment, from month to month, but not significantly. The variations were not always in the ascending order as weights of pasture-fed animals, even those which had not passed the age of maturity, were greatly influenced by the stand of pasture grass in different months of the year. Two of the females calved during the progress of the experiment. As may be expected, the males were heavier than the females. The average weight of the males during the period of the experiment was 300.5 kgm. whereas the females averaged 209.5 kgm.

Hemoglobin indices

The data obtained on the hemoglobin determinations at intervals of one week during the entire period of the experiment show that the hemoglobin readings in all animals from month to month were rather variable; the coefficients of variability oscillating from 2.5 to 39.7 per cent. The greatest variability was observed during the month of July. The trend of fluctuations of hemoglobin readings being fairly similar in all individuals, a description of one of them will suffice to show the fluctuations for the group. Thus, for the steer named Magno, No. 280, the monthly mean of hemoglobin was 8.92 $\pm$ 0.303 grams per 100 ml. of blood for January, 8.48 $\pm$ 0.159 grams for February, 8.38 $\pm$ 0.149 grams for March, 7.85 $\pm$ 0.201 grams for April, 7.91 $\pm$ 0.195 grams for May, 7.95 $\pm$ 0.275 grams for June, 9.19 $\pm$ 0.441 grams for July, 9.35 $\pm$ 0.138 grams for August, 8.95 $\pm$ 0.177 grams for September, 9.45 $\pm$ 0.336 grams for October, 11.15 $\pm$ 0.328 grams for November, and 11.29 $\pm$ 0.449 grams for December.

To test whether the hemoglobin of the males at different atmospheric temperature levels was different from those of the females, Snedecor's (1937) analysis of variance was employed. In none of the different atmospheric temperature levels was the value of F greater than one, thus showing that the hemoglobin of the males was not different from those of the females.

On the whole, the higher values for hemoglobin occurred in the cooler months of the year. The average for the year was 8.75 grams per 100 ml. of blood (table 1). This average is lower by 0.68 gram per 100 ml. than that reported by Manresa and Reyes (1934) for the same breed of cattle, namely: 9.43 ± 0.13 grams per 100 ml. of blood. Manresa and Falcon (1939) reported the hemoglobin index for Indian Nellore cattle taken at 2 p. m. throughout the entire year to be 9.79 grams per 100 ml.

Zialcita [2] found the index for an Indian Nellore bull taken at the same time of the day to be 8.44 grams per 100 ml. The difference between these determinations is greater than the difference between

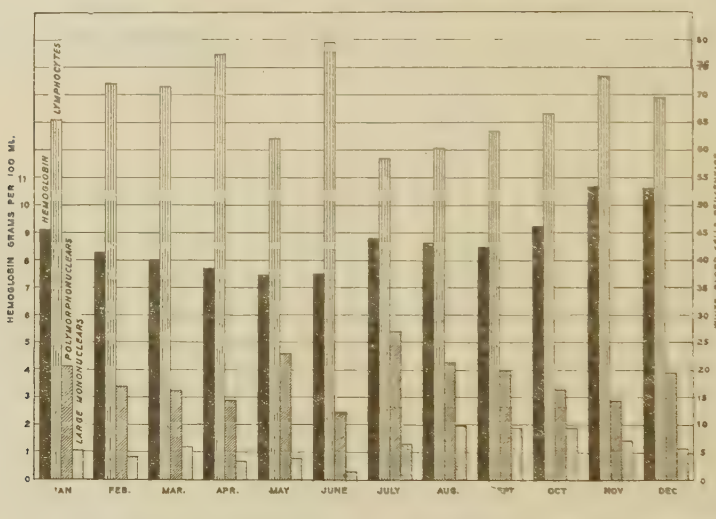

Fig. 1.—Monthly fluctuation of hemoglobin and differential white blood cell counts for lymphocytes, large mononuclears and polymorphonuclears in Philippine Native cattle.

the present data and those reported by Manresa and Reyes (1934). The hemoglobin determinations obtained by Manresa, Gomez, and Santos (1939) in mature Nellore cattle during a course of twenty-four hours for a period of seven days have also shown that the hemoglobin indices were higher during the cooler part of the day; that is, in the morning and late in the afternoon, when the atmospheric temperature was relatively low; and towards noon, when the temperature of the air was relatively high, the hemoglobin of the animal was low.

[2] ZIALCITA, L. P. A comparative study of the blood in cattle in relation to constitutional vigor. (Thesis presented for graduation, 1938, with the degree of Bachelor of Science in Agriculture from the College of Agriculture. Unpublished).

As shown in figures 1 and 2, the fluctuations of the hemoglobin indices for both males and females may be described as double cycled. The first cycle is of seven months duration, starting in October and ending in April, with the peak in the month of November. The second cycle is of five months duration starting in May and ending in September, with the mode in July. These trends in the yearly fluctuations in hemoglobin for Philippine Native cattle are in close agreement with those obtained by Manresa and Falcon (1939) for Indian Nellore cattle.

Body temperature

The average body temperature of all animals during the experiment was 39.10°C. This average is higher than that obtained

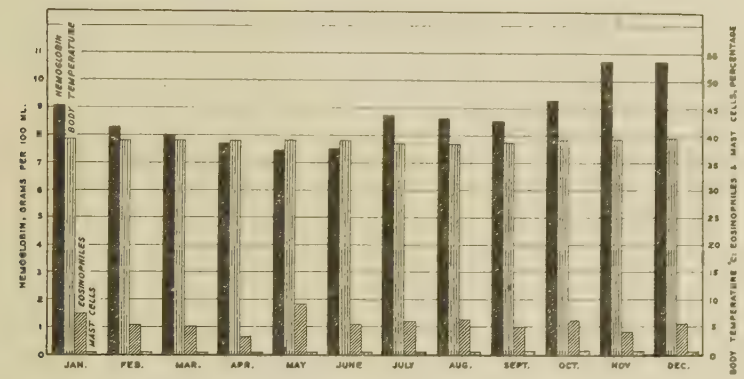

Fig. 2.—Monthly fluctuation of hemoglobin, body temperature, and differential white blood cell counts for eosinophiles and mast cells in Philippine Native cattle.

by Manresa and Falcon (1939), 38.89°C., for Indian Nellore cattle. The correlations of the monthly fluctuation of hemoglobin with body temperature were (r) 0.20, 0.14, 0.29, 0.09, 0.27, and 0.17 for animals Nos. 280, 284, 285, 275, 277, and 283, respectively. All the correlations were positive; however, none of them was significant even at 5 per cent level for 10 degrees of freedom. It may be safely said that the body temperature did not enter as a factor in the changes of hemoglobin.

In the warm months of the year, from February to August, 1939, the mean of body temperature was 39.12 ± 0.029°C., whereas in the cooler months of the year, from September to January, the mean was 39.04 ± 0.037 °C. The difference of the means

of 0.08 ± 0.047°C. was not significant. Since the average atmospheric temperature during the progress of the experiment was 31.3°C. for the warm months and 27.4°C. for the cooler months, it seems clear that the six head of Philippine Native cattle studied were not greatly affected by the changes in atmospheric temperature.

In an experiment with heavy milking cows belonging to the Holstein-Friesian, Jersey, and Guernsey breeds of cattle which were housed for varying lengths of time in a large air-conditioned room in which the temperature was increased from 40°F. to 100°F. (4.4 to 37.7°C.) at intervals of 10°F., and with relative humidity and air movement remaining constant, Regan and Richardson (1938) found that as the room temperature increased, the body temperature also increased uniformly up to a point where the animals were no longer able to maintain heat balance. The body temperature of European cattle raised in the tropics such as certain regions of Brazil was found by Rhoad (1936) to rise readily with an increase of external temperature. Indian breeds, however, reacted but slightly, thus confirming our findings.

Manresa and Erce (1940), working on a large number of Holstein-Friesian and Jersey cows at the Hacarin Dairy Farm, San José del Monte, Bulacan, found the average body temperature of these cows from 2 to 4 p. m. to be significantly higher than the average from 6 to 8 a.m. In temperate regions where European cattle have become acclimated, Regan and Richardson (1938) stated that the Holstein, Jersey, and Guernsey cows withstand long periods of exposure to temperature below zero degrees Fahrenheit with little loss either in milk production or in their efficiency to utilize feed. On the other hand, Rhoad (1936) stated that short periods of extreme thermal condition may be supported by most warm-blooded animals without endangering health or bodily functions, but that prolonged periods of the same abnormal condition may seriously affect their health and production, inasmuch as the animal organism tends to adjust itself to a new environment to the extent it is capable. At the Iberian Livestock Experiment Farm at Jeanerette, Louisiana, Rhoad (1938) found that the body temperature of the Aberdeen-Angus (*Bos taurus*) was influenced to a greater degree by a high atmospheric temperature than that of the Guzerat breed of cattle (*Bos indicus*). When exposed to direct solar radiation during summer for a considerable length of time, a febrile condition in the *Bos taurus* was noted, whereas in the *Bos indicus* this was not

true. Rhoad (1938) further found that the body temperature of the purebred and that of the half-bred Brahman were higher than those of the purebred and the quarterbred Aberdeen-Angus at low temperature. It was also found that the body temperature of the purebred Brahman (*Bos indicus*) decreased with rising external temperature until a heat balance was established, after which the body temperature rose.

In this connection, it should be remembered that the southernmost portion of the United States enjoys a sub-tropical climate warmer than the northern states of the United States. The atmospheric temperature in Louisiana ranges, according to Davidson (1927), from 59°F. to 81°F. (15.0°C. to 27.2°C.), and the relative humidity is 58 per cent. This was pointed out by Davidson as the reason that in this state large breeds of dairy cattle like the Holstein-Friesian could not be raised.

Correlation between hemoglobin and relative humidity

In the experiment of Manresa and Falcon (1939), the difference of average relative humidity prevailing during the cool and warm months of the year was only 2.7 per cent. They found a low positive correlation between hemoglobin and relative humidity. In the present experiment, the relative humidity prevailing during the warm months of the year, namely, from March to August was found to be lower than that prevailing during the cool months of the year. This partly accounts for the moderately high positive correlation coefficient of 0.404 ± 0.032 between hemoglobin and relative humidity. Of the factors which may have helped to increase the degree of correlation, the availability of feed supply must be mentioned. Manresa and Reyes (1934) found that the higher hemoglobin indices on the whole occurred in animals of excellent physical condition. Since the animals in both experiments were pasture-fed, it can only be surmised that the Philippine Native cattle do not fare so poorly on pasture grass alone.

Relationship between the hemoglobin and the different kinds of white blood corpuscles

A summary of the data on differential white blood cell counts for males and females and the combined data for males and females are presented in table 2.

Lymphocytes. The percentages of lymphocytes presented in table 2 varied distinctly from month to month. The mean for the males was 69.50 ± 0.621 per cent and for the females, 66.67 ± 0.553 per cent. The difference in the percentage of lymphocytes between the males and the females was found to be significant. The mean for both males and females was 67.84 ± 0.594 per cent. This percentage is very much higher than that reported by Manresa and Falcon (1939), which was 54.20 per cent in Indian Nellore oxen.

Large mononuclears. In table 2, the percentages of large mononuclears in both males and females can be seen to vary greatly from month to month. The mean percentage for males was 6.05 ± 0.499, and for females, 6.38 ± 0.513 per cent. The mean for all male and female animals was 6.21 ± 0.499. The difference in the percentage of large mononuclears between the males and the females is not significant. This percentage is higher than that reported by Manresa and Falcon (1939), which was 3.51 ± 0.050, and that found by Zialcita [3], which was 4.45 per cent.

Polymorphonuclears. The mean monthly percentage of polymorphonuclears in both males and females was 17.38 ± 0.438 for the males, and 19.05 ± 0.375 for the females. The difference of 1.67 ± 0.594 per cent is not significant. The mean for both males and females was 18.23 ± 0.412 per cent. This is lower than the average, 30.5 per cent, reported by Dimock and Thompson in 1906 (Burnett, 1917) for normal cattle in the United States. Zialcita [4] working on Holstein-Friesian ox raised in the Philippines found the differential polymorphonuclears to be 26.28 per cent.

Eosinophiles. The percentages of eosinophiles in the blood of Philippine Native cattle varied greatly from month to month. The mean for the males was 5.55 ± 0.286 per cent, and for the females, 6.22 ± 0.340. The mean for all the animals was 6.05 ± 0.281. This is lower than the result obtained by Manresa and Falcon (1939) and Zialcita [5], but the variations were within the maximum and minimum limits of the findings of Dimock and Thompson in 1906 (Burnett, 1917).

[3,4,5] See footnote 2.

Mast cells. The percentage of mast cells did not vary greatly from month to month during the whole period of the experiment. In the males, the mean was 0.43 ± 0.376; and in the females, 0.59 ± 0.322 per cent. The mean average for both males and females was 0.520 ± 0.302. This figure is slightly lower than that reported by Manresa and Falcon (1939), who found the percentage for Indian Nellore cattle to be 0.78 ± 0.018. Zialcita [6] reported a percentage of 0.85 from a Nellore bull. According to Burnett (1917), Dimock and Thompson in 1906 obtained an average percentage of 0.59 of mast cells for normal cattle.

As shown in figure 1, no consistent agreement in the fluctuations of the hemoglobin occurred among the three varieties of white blood cells. The outstanding difference between the data here reported and those of Manresa and Falcon (1939) is the fact that the percentage of lymphocytes in Philippine Native cattle stood far above that of the Indian Nellore throughout the experiment. The mean for the Philippine Native cattle for the year was 67.84 ± 0.594 per cent and that for the Nellore, 51.31 ± 0.203. In the Indian Nellore, the coefficient of variation was 1.95 ± 0.268 per cent, and in the Philippine Native cattle, 4.49 ± 0.618 per cent. This shows that these cells were more variable in the Philippine Native than in the Nellore. The same general statement as regards the variability of polymorphonuclears and large mononuclears can be made for Philippine Native cattle when compared to the Indian Nellore. It appears that the variation in percentages of white blood cells is general not only for Philippine animals but also for cattle in temperate regions as substantiated by Dimock and Thompson in 1906 (Burnett, 1917) and by Burnett (1917).

The mean monthly percentage of eosinophiles, as shown in table 2, was 6.05 ± 0.281 and that of mast cells, 0.52 ± 0.302. Graphical presentation of the relationship between these varieties of leucocytes with hemoglobin is shown in figure 2. No agreement was found between the eosinophiles and mast cells with hemoglobin. Manresa and Falcon (1939) obtained the same results in Indian Nellore cattle.

[6] See footnote 2.

A summary of the data on leucocytes in this study is tabulated below. The white blood cell counts reported by Manresa and Falcon (1939) and by Dimock and Thompson in 1906 (Burnett, 1917) are included in the tabulation for purposes of comparison.

The differential white blood cell counts in per cent in Philippine native cattle

VARIETIES OF CELLS	DATA FROM PRESENT WORK			MANRESA AND FALCON (1939)			DIMOCK AND THOMPSON (1906)		
	Min.	Max.	Mean	Min.	Max.	Av.	Min.	Max.	Av.
Lymphocytes ...	58.99	79.58	67.84∓0.594	43.13	61.22	51.31	31.00	76.00	54.20
Large mononuclears	1.74	10.01	6.21∓0.499	1.28	6.22	3.51	0.20	3.30	1.40
Polymorphonuclears	12.43	27.22	18.23∓0.412	21.16	41.51	30.18	13.00	43.80	30.50
Eosinophiles	3.44	9.66	6.05∓0.281	6.17	20.93	14.22	3.80	26.50	13.15
Mast cells	0.27	0.87	0.52∓0.302	0.0	1.65	0.78	0.10	1.20	0.59

SUMMARY

1. A hematological study conducted for a period of one year on three male and three female Philippine Native cattle showed that the average hemoglobin indices for the six animals of both sexes studied of this breed were 8.75 grams per 100 ml. of blood.

2. The fluctuations of hemoglobin indices for both the male and female oxen for the entire year was two-cyclic. The first cycle was of seven months duration, starting in October and ending in April; and the second cycle lasted for five months, starting in May and ending in September.

3. The average body temperature for the entire period of the experiment was 39.10 °C. No differences were found between the average body temperature during the cool months of the year and those of the warm months.

4. The mean percentages of the different varieties of leucocytes were as follows: lymphocytes, 67.84; large mononuclears, 6.21; polymorphonuclears, 18.23; eosinophiles, 6.05; and mast cells, 0.52.

5. No consistent relationship was found between the normal fluctuation in hemoglobin indices and the percentages of lymphocytes, large mononuclears, polymorphonuclears, eosinophiles, and mast cells.

6. No correlation was found between hemoglobin and body temperature, but the hemoglobin was negatively correlated with atmospheric temperature.

7. The body temperature of Philippine Native cattle during the warm months of the year under average atmospheric temperature 31.3°C. was not significantly higher than the body temperature during the cooler months of the year under average atmospheric temperature 27.4°C. This is contrary to the findings in the Jersey and Holstein Friesian breeds of dairy cattle reared in the Philippines. It is probable that the ability to withstand external heat without significantly raising the temperature of the body might be used as a test for adaptability of different breeds of cattle to a given environment.

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TABLE 1

A summary of the physical examination of the blood of six Philippine native cattle from July 16, 1938 to July 15, 1939

NAMES OF ANIMALS	MONTHS	WEIGHT	HEMOGLOBIN INDEX; GRAMS PER 100 ML.	BODY TEMPERATURE	ATMOSPHERIC TEMPERATURE	HUMIDITY
		kgm.	*grams*	*°C.*	*°C.*	*per cent*
Magno No. 280	January	256.4	8.92	39.08	25.90	61.75
	February	286.4	8.48	38.92	27.07	56.75
	March	299.1	8.38	39.05	31.60	40.25
	April	320.0	7.85	39.02	31.40	47.20
	May	306.4	7.91	39.06	31.42	58.75
	June	319.1	7.95	39.16	31.20	58.75
	July	283.6	9.19	38.93	30.54	59.40
	August	310.0	9.35	38.69	31.67	53.00
	September	295.0	8.95	39.00	29.35	67.00
	October	304.5	9.45	39.51	27.90	71.60
	November	305.5	11.15	39.33	28.05	62.25
	December	309.1	11.29	39.24	26.42	69.00
	Average	299.6	9.07	39.08	29.37	58.80
Sofronio No. 285	January	262.7	8.72	38.94	25.90	61.75
	February	255.5	8.12	39.08	27.07	56.75
	March	259.1	8.35	38.89	31.60	40.25
	April	282.7	7.89	38.93	31.40	47.20
	May	274.5	7.71	39.03	31.42	58.75
	June	280.0	7.98	38.89	31.20	58.75
	July	255.5	9.21	38.80	30.54	59.40
	August	277.3	8.61	38.47	31.67	53.00
	September	261.4	8.45	38.61	29.35	67.00
	October	272.7	9.11	39.11	27.90	71.00
	November	268.2	10.78	39.03	28.05	62.25
	December	277.3	10.58	39.24	26.42	69.00
	Average	268.9	8.79	38.92	29.37	58.80
Silvestre No. 284	January	336.4	8.35	39.06	25.90	61.75
	February	340.9	8.25	39.16	27.07	56.75
	March	340.0	8.05	39.03	31.60	40.25
	April	357.3	7.56	38.96	31.40	47.20
	May	247.3	7.45	38.95	31.42	58.75
	June	366.4	7.75	39.11	31.20	58.75
	July	320.9	8.52	38.93	30.54	59.40
	August	318.2	8.12	38.64	31.67	53.00
	September	328.2	8.35	38.81	29.35	67.00
	October	340.9	8.82	39.28	27.90	71.60
	November	350.9	10.71	39.05	28.05	62.25
	December	347.3	10.68	39.13	26.42	69.00
	Average	332.9	8.55	39.01	29.37	58.80
Average for male		300.5	8.80	39.00	29.37	58.80

TABLE 1 (Continued)

NAMES OF ANIMALS	MONTHS	WEIGHT	HEMOGLOBIN INDEX; GRAMS PER 100 ML.	BODY TEMPERATURE	ATMOSPHERIC TEMPERATURE	HUMIDITY
		kgm.	*grams*	°C.	°C.	*per cent*
Maxima No. 275	January	212.7	9.58	39.22	25.90	61.75
	February	166.4	7.62	39.35	27.07	56.75
	March	172.7	7.08	39.27	31.60	40.25
	April	156.4	6.92	39.09	31.40	47.20
	May	156.4	6.85	39.30	31.42	58.75
	June	163.6	6.92	39.33	31.20	58.75
	July	190.9	8.55	39.15	30.54	59.40
	August	190.9	9.75	38.78	31.67	53.00
	September	195.5	8.95	38.94	29.35	67.00
	October	204.5	9.38	39.85	27.90	71.60
	November	208.2	10.12	39.61	28.05	62.25
	December	173.6	10.17	39.42	26.42	69.00
	Average	182.7	8.49	39.27	29.37	58.80
Marta No. 277	January	198.2	9.71	39.53	25.90	61.75
	February	203.6	8.71	39.33	27.07	56.75
	March	197.3	8.45	39.63	31.60	40.25
	April	200.9	7.94	39.37	31.40	47.20
	May	200.0	8.25	39.58	31.42	58.75
	June	201.8	8.08	39.58	31.20	58.75
	July	185.5	8.65	39.24	30.54	59.40
	August	178.2	7.35	38.68	31.67	53.00
	September	174.5	7.85	39.11	29.35	67.00
	October	174.5	9.35	39.88	27.90	71.60
	November	187.3	10.55	39.63	28.05	62.25
	December	176.4	10.23	39.75	26.42	69.00
	Average	189.4	8.76	39.44	29.37	58.80
Sampaguita No. 283	January	290.0	9.35	38.78	25.90	61.75
	February	300.9	8.75	39.03	27.07	56.75
	March	254.5	8.25	38.72	31.60	40.25
	April	231.8	8.15	39.02	31.40	47.20
	May	230.9	7.12	39.33	31.42	58.75
	June	238.2	6.80	39.11	31.20	58.75
	July	278.2	8.74	38.82	30.54	59.40
	August	270.9	8.62	38.47	31.67	53.00
	September	259.1	8.62	38.58	29.35	67.00
	October	259.1	9.56	39.11	27.90	71.60
	November	280.9	10.97	38.98	28.05	62.25
	December	176.4	11.22	38.99	26.42	69.00
	Average	255.9	8.84	38.91	29.37	58.80
Average for female		209.5	8.69	39.206	29.37	58.80
General average		255.0	8.75	39.103	29.37	58.80

TABLE 2

Monthly averages of the differential white blood cell counts from July 16, 1938 to July 15, 1939

Males

NAMES OF CELLS	JANUARY	FEBRUARY	MARCH	APRIL	MAY	JUNE	JULY	AUGUST
Lymphocytes	66.04	72.43	74.97	79.70	64.11	81.05	58.12	63.30
Large mononuclears ..	4.64	4.60	6.05	3.77	4.72	1.53	6.99	9.48
Polymorphonuclears ..	22.19	17.05	13.63	12.97	21.73	11.48	28.44	20.70
Eosinophiles	6.92	5.14	4.95	3.12	9.14	5.49	5.85	5.77
Mast cells	0.21	0.78	0.40	0.44	0.30	0.45	0.60	0.75

NAMES OF CELLS	SEPTEMBER	OCTOBER	NOVEMBER	DECEMBER	AVERAGES	S. D.	C. V.
Lymphocytes	65.67	66.48	75.26	70.75	69.50 ± 0.621	3.188 ± 0.439	4.587 ± 0.632
Large mononuclears ..	10.03	10.29	6.37	5.38	6.05 ± 0.499	2.565 ± 0.353	42.390 ± 6.800
Polymorphonuclears ..	8.35	16.60	14.27	18.51	17.38 ± 0.438	2.251 ± 0.309	12.948 ± 1.807
Eosinophiles	5.43	6.05	3.88	5.25	5.55 ± 0.286	1.471 ± 0.203	26.504 ± 3.868
Mast cells	0.52	0.58	0.22	0.11	0.43 ± 0.376	1.932 ± 0.376	45.920 ± 6.323

TABLE 2 (continued)

Females

NAMES OF CELLS	JANUARY	FEBRUARY	MARCH	APRIL	MAY	JUNE	JULY	AUGUST
Lymphocytes	65.36	71.96	68.67	75.58	60.32	78.11	59.87	58.35
Large mononuclears ..	6.36	4.45	6.02	3.99	4.16	1.94	6.78	10.54
Polymorphonuclears ..	19.79	17.19	19.18	16.19	24.85	13.38	26.00	22.58
Eosinophiles	7.98	5.69	5.81	3.75	10.18	6.19	6.78	7.53
Mast cells	0.51	0.71	0.32	0.49	0.49	0.38	0.57	1.00

NAMES OF CELLS	SEPTEMBER	OCTOBER	NOVEMBER	DECEMBER	AVERAGES	S. D.	C. V.
Lymphocytes	62.00	66.53	71.66	67.51	66.67 ± 0.553	2.842 ± 0.391	4.262 ± 0.587
Large mononuclears ..	9.82	9.39	8.07	5.89	6.38 ± 0.513	2.634 ± 0.363	41.285 ± 6.577
Polymorphonuclears ..	22.35	16.66	15.24	20.39	19.05 ± 0.375	1.925 ± 0.265	10.104 ± 1.404
Eosinophiles	5.08	6.71	4.33	5.79	6.22 ± 0.340	1.748 ± 0.241	28.102 ± 4.147
Mast cells	0.75	0.71	0.70	0.42	0.59 ± 0.322	1.656 ± 0.228	28.060 ± 4.095

TABLE 2 (continued)

Averages for males and females

NAMES OF CELLS	JANUARY	FEBRUARY	MARCH	APRIL	MAY	JUNE	JULY	AUGUST
Lymphocytes	65.70	72.19	71.82	77.64	62.22	79.58	58.99	60.83
Large mononuclears ..	5.50	4.53	6.03	3.88	4.44	1.74	6.89	10.01
Polymorphonuclears ..	20.99	17.12	16.41	14.58	23.29	12.43	27.22	21.64
Eosinophiles	7.45	5.42	5.38	3.44	9.66	5.84	6.31	6.65
Mast cells	0.36	0.74	0.36	0.46	0.39	0.41	0.59	0.87

NAMES OF CELLS	SEPTEMBER	OCTOBER	NOVEMBER	DECEMBER	AVERAGES	S. D.	C. V.
Lymphocytes	63.83	66.51	73.46	69.13	67.84 $\pm$ 0.594	3.052 $\pm$ 0.420	4.490 $\pm$ 0.618
Large mononuclears ..	9.93	9.84	7.22	5.63	6.21 $\pm$ 0.499	2.563 $\pm$ 0.352	41.270 $\pm$ 6.575
Polymorphonuclears ..	20.35	16.63	14.75	19.45	18.23 $\pm$ 0.412	2.114 $\pm$ 0.291	11.590 $\pm$ 1.611
Eosinophiles	5.25	6.38	4.11	5.52	6.05 $\pm$ 0.281	1.442 $\pm$ 0.198	23.830 $\pm$ 3.571
Mast cells	0.64	0.64	0.46	0.27	0.52 $\pm$ 0.302	1.554 $\pm$ 0.213	30.000 $\pm$ 3.826

ABSORPTION OF SOIL POTASSIUM BY RICE[1]

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Of the Department of Soils

WITH TWO TEXT FIGURES

The present study was undertaken to secure additional information as to the time when rice grown on upland soils needs an ample supply of potassium. The data may serve also as a guide for increasing the producing capacity of upland soil for rice.

The importance of potassium has attracted the attention of investigators, especially Kelly and Thompson (1910), Gile and Carrero (1915), Burd (1919), and Remy (1931).

MATERIALS AND METHODS

The rice grains used in this study were those of the Inintiw variety secured from the Department of Agronomy. The grains contained 0.146 per cent CaO, 0.282 per cent MgO, and 0.432 per cent K_2O. Because this variety matures in a much shorter time than other varieties, it is generally planted in upland soil and in localities where the water supply is very limited and the farmers depend upon rain for their irrigation.

Lipa clay loam collected near the Rural High School site, College of Agriculture, was used. Its colloid content was 40.38 per cent; water-holding capacity, 52.80 per cent; plant foods held in reserve, SiO_2–0.45 per cent; TiO_2–0.46 per cent; Al_2O_3–5.25 per cent; Fe_2O_3–8.35 per cent; Mn_3O_4–0.18 per cent; CaO–0.43 per cent; MgO–0.87 per cent; K_2O–0.18 per cent; Na_2O–0.21 per cent; P_2O_5–0.22 per cent; SO_3–0.18 per cent; N–0.16 per cent; and available nutrients, K_2O–0.03 per cent; P_2O_5–0.01 per cent.

Ninety-eight kerosene cans, cut in halves, 9.5″ × 9.5″ × 7.0″ and with the inner and outer sides coated with coal tar were used as pots for the experiments.

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[1] Experiment Station contribution No. 1438.

Eight kilograms of soil were placed in each pot, watered with two-thirds of its water-holding capacity, and planted to palay seeds previously soaked in water for 24 hours.

The cultures which consisted of fourteen lots of seven replications each were exposed to the sun and rain, given sufficient water, weeded, and cultivated from time to time as needed to make the soil conditions optimum for plant development. It was observed that the plants grown during the wet season were apparently more vigorous, taller and darker, and likewise had more tillers and broader leaves than those in the dry season. In the preparation of the ash extracts, it was observed that the extracts of older plants were more intensely colored yellow than those of the younger ones and thus showed that more iron was absorbed from the soil as the plants became older.

The first lot was harvested two weeks after planting while the remaining thirteen lots, at intervals of one week. The harvest which consisted of the portion of the plant above the ground and the rootlets were carefully freed from soil particles, dried, and then ashed in the usual way.

The potassium content of the ash was determined in accordance with the Sherrill centrifugal method as prescribed by the Association of Hawaiian Sugar Technologists (1931). The amount of CaO and MgO were determined with the use of the method recommended by the Association of Agricultural Chemists (1935). The amount of soil minerals, K_2O, CaO, and MgO absorbed by the rice plants was determined by substracting the mineral, K_2O, CaO, and MgO contents of the rice seeds from those of the entire plants.

RESULTS AND DISCUSSION

The results of the different determinations are given in table I and graphically shown in figures 1 and 2.

It is evident in the data given in table 1 that as the rice plants in both the dry and wet season cultures became older, the amount of minerals absorbed from the soil increased (fig. 2), whereas that of K_2O, CaO, and MgO decreased (fig. 1). This decrease with age may be explained by the fact that before the plants reached maturity, the inorganic bases remigrated to the soil by the excretion of the roots, cuticular sloughing of the living tissues of the plants, and lixiviation of the dead and decaying tissues. In both cultures, the decrease in the amounts of calcium and magnesium

was gradual, whereas that of potash was abrupt, as may be seen in figure 1.

These findings point out that in soil fertility work, in connection with rice plants cultivated on upland soils, the essential soil base-forming elements should be made available during their early stage of growth. The addition, therefore, of fertilizers which may increase the amount of available soil potash as well as liming mate-

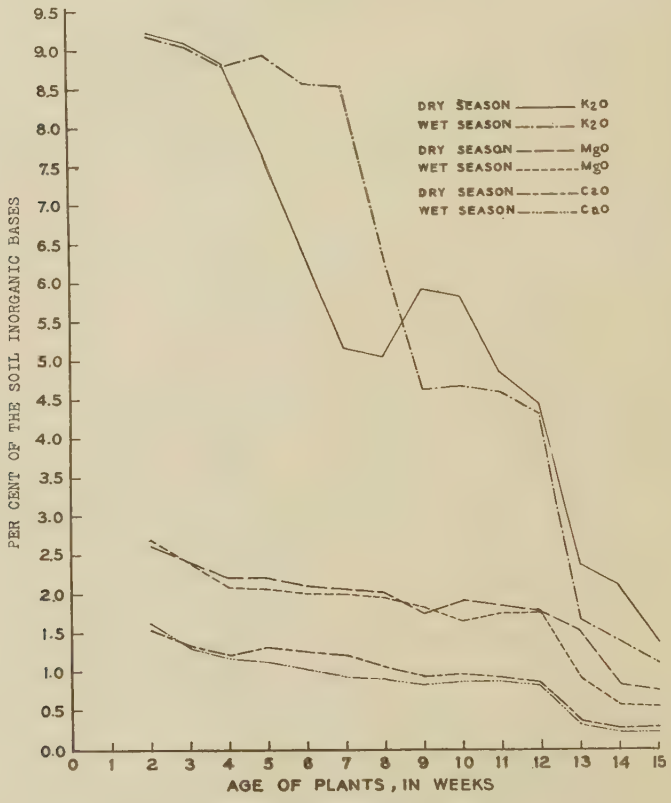

Fig. 1.—Distribution of the soil inorganic bases at the different stages of growth of rice plants.

rials should be made during the vegetative growth of the plants rather than during the age of maturation. On the other hand, the other constituents of the ash of the plants should be present in the soil in a comparatively greater amount during the age of maturity. It is also apparent in this table that of the three soil bases absorbed by the plants, the amount of potash appeared to be the highest and that of CaO, the lowest. These results are revealed by both the

dry and wet season cultures. It may indicate further that in the addition of inorganic bases to soils intended for growing rice, relatively higher amount of potash than either calcium or magnesium should be applied. It may also be assumed that potassium is the most essential element for rice, with calcium and magnesium as equally important. This assertion is substantiated by the investigation carried out by Newton (1928) who reported that wheat,

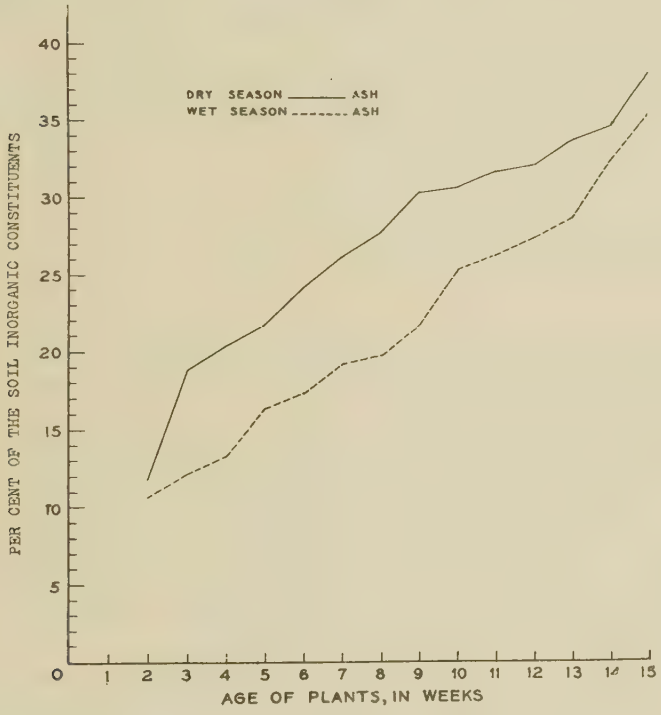

Fig. 2.—Distribution of the soil inorganic constituents at different stages of growth of rice plants.

which like rice belongs to the Gramineae, absorbed comparatively more K_2O, than either CaO or MgO when grown in loam soil.

When the data of the dry season cultures are compared with those of the wet season, it is obvious that the dry season cultures had a tendency to absorb a higher amount of soil inorganic constituents and bases, namely, K_2O, CaO, and MgO, than the wet season cultures particularly towards maturity (figs. 1 and 2). This result may induce one to assert that the amount of precipitation influences the rate of absorption. This phenomenon may be attributed

to the fact that in season of greater rainfall, the available constituents of soils exist in a relatively more dilute concentration than those in the dry season.

In order to have an idea of the degree in which the inorganic constituents of the rice plants are associated with each other, the coefficients of correlation are calculated and given in table 2. It is apparent in this table that the values of the coefficient of correlation ($\sqrt{}$) between the inorganic constituents and the three bases, K_2O, CaO, and MgO, are all negative and highly significant in both the dry and wet season cultures. This tends to point out that, with age, the increase of the ash content of rice plants is accompanied by a decrease of K_2O, CaO, and MgO. Positive and highly significant correlations are shown by the amounts of K_2O, CaO, and MgO. This indicates that an increase of one of the bases in the body of the plants means also an increase of the other two.

SUMMARY

1. The rice plants in the wet-season cultures were bigger, darker, taller, and had more tillers than those of the dry season.

2. The percentage of the inorganic constituents increased with age whereas that of K_2O, CaO, and MgO decreased. The decrease in the amount of K_2O absorbed from the soil by rice plants was seemingly abrupt as compared to that of CaO and MgO. Absorption of these soil bases was greatest during the seedling stage and least at maturity which indicates that they should be made available when the rice plants are in their seedling stage.

3. The absorption of the inorganic soil constituents by the plants during the dry season was relatively higher than during the wet season.

4. Of the soil bases absorbed by rice plants from the soil, potassium was the highest. Positive and significant coefficient of correlation was shown by the amount of soil bases which were absorbed by the rice plants. This correlation indicated that the trend of absorption for soil-K_2O,-CaO, and-MgO is parallel and similar.

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TABLE 1

Percentage of soil inorganic constituents absorbed by the rice plants at different stages of growth

Dry season culture

(Rainfall = .03 — 53.83 mm.) [a]
(Temperature = 34.5° — 29.5°C)

LOT NUMBER	DATE HARVESTED	INORGANIC CONSTITUENTS	K_2O	CaO	MgO
		Mean	Mean	Mean	Mean
		per cent	*per cent*	*per cent*	*per cent*
I	25— I—1940	11.93 ∓ 0.337	9.33 ∓ 0.033	1.56 ∓ 0.004	2.63 ∓ 0.010
II	1— II—1940	18.62 ∓ 0.081	9.10 ∓ 0.032	1.34 ∓ 0.014	2.42 ∓ 0.083
III	8— II—1940	20.46 ∓ 0.113	8.80 ∓ 0.176	1.21 ∓ 0.014	2.10 ∓ 0.070
IV	15— II—1940	21.74 ∓ 0.084	7.60 ∓ 0.059	1.33 ∓ 0.008	2.22 ∓ 0.008
V	22— II—1940	24.01 ∓ 0.470	6.33 ∓ 0.029	1.29 ∓ 0.002	2.11 ∓ 0.004
VI	29— II—1940	26.11 ∓ 0.489	5.17 ∓ 0.089	1.22 ∓ 0.007	2.08 ∓ 0.046
VII	7— IV—1940	27.74 ∓ 0.310	5.01 ∓ 0.157	1.07 ∓ 0.028	2.02 ∓ 0.009
VIII	14— III—1940	30.25 ∓ 0.493	5.85 ∓ 0.037	0.91 ∓ 0.016	1.79 ∓ 0.023
IX	21— III—1940	31.04 ∓ 0.423	5.83 ∓ 0.051	0.95 ∓ 0.021	1.92 ∓ 0.007
X	28— III—1940	31.79 ∓ 1.290	4.87 ∓ 0.059	0.92 ∓ 0.032	1.84 ∓ 0.007
XI	4— IV—1940	31.83 ∓ 0.552	4.44 ∓ 0.066	0.89 ∓ 0.029	1.80 ∓ 0.230
XII	11— IV—1940	33.76 ∓ 0.543	2.30 ∓ 0.045	0.38 ∓ 0.006	1.01 ∓ 0.069
XIII	18— IV—1940	34.79 ∓ 1.139	2.05 ∓ 0.051	0.28 ∓ 0.039	0.85 ∓ 0.078
XIV	25— IV—1940	37.52 ∓ 0.940	1.35 ∓ 0.006	0.27 ∓ 0.010	0.71 ∓ 0.011

[a] Ranges of rainfall and temperatures from January 25, to April 25, 1940, taken from the Department of Agricultural Botany, College of Agriculture.

TABLE 1 (Continued)

Wet season culture *(Rainfall = 0.5 — 71 mm)* [b] *(Temperature = 43.5° — 27°C.)*

LOT NUMBER	DATE HARVESTED	INORGANIC CONSTITUENTS	K_2O	CaO	MgO
		Mean	Mean	Mean	Mean
I	6— VI—1940	10.86 ∓ 0.195	9.22 ∓ 0.037	1.63 ∓ 0.284	2.70 ∓ 0.035
II	20— VI—1940	12.07 ∓ 0.171	9.05 ∓ 0.026	1.32 ∓ 0.012	2.41 ∓ 0.013
III	27— VI—1940	13.35 ∓ 0.011	8.77 ∓ 0.521	1.22 ∓ 0.042	2.24 ∓ 0.039
IV	4— VII—1940	16.29 ∓ 0.062	8.92 ∓ 0.030	1.14 ∓ 0.028	2.13 ∓ 0.025
V	11— VII—1940	17.39 ∓ 0.047	8.57 ∓ 0.001	1.02 ∓ 0.027	2.01 ∓ 0.046
VI	18— VII—1940	19.08 ∓ 0.046	8.51 ∓ 0.001	0.95 ∓ 0.008	1.98 ∓ 0.035
VII	25— VII—1940	19.93 ∓ 0.283	6.38 ∓ 0.035	0.90 ∓ 0.028	1.85 ∓ 0.039
VIII	1—VIII—1940	21.64 ∓ 0.315	4.57 ∓ 0.774	0.84 ∓ 0.030	1.67 ∓ 0.046
IX	8—VIII—1940	25.41 ∓ 0.164	4.65 ∓ 0.006	0.88 ∓ 0.024	1.75 ∓ 0.009
X	15—VIII—1940	26.37 ∓ 0.270	4.50 ∓ 0.034	0.88 ∓ 0.018	1.77 ∓ 0.016
XI	22—VIII—1940	27.35 ∓ 0.402	4.43 ∓ 0.152	0.85 ∓ 0.007	1.72 ∓ 0.136
XII	29—VIII—1940	28.69 ∓ 0.010	1.63 ∓ 0.010	0.43 ∓ 0.024	0.95 ∓ 0.043
XIII	5— IX—1940	32.39 ∓ 0.030	1.38 ∓ 0.006	0.26 ∓ 0.022	0.51 ∓ 0.026
XIV	12— IX—1940	34.43 ∓ 0.611	1.01 ∓ 0.009	0.20 ∓ 0.010	0.46 ∓ 0.007

[b] Ranges of rainfall and temperatures from June 6 to September 12, 1940, obtained from the Department of Agricultural Botany, College of Agriculture.

TABLE 2

Coefficient of correlation between the inorganic constituents of rice plants

CONSTITUENTS CORRELATED	VALUE OF COEFFICIENT OF CORRELATION—r	
	Dry season	Wet season
	per cent	*per cent*
Inorganic constituents—K_2O	— 0.931	— 0.947
Inorganic constituents—CaO	— 0.619	— 0.930
Inorganic constituents—MgO	— 0.679	— 0.924
K_2O — CaO	+ 0.917	+ 0.907
K_2O — MgO	+ 0.926	+ 0.935
CaO — MgO	+ 0.533	+ 0.933

STUDIES ON THE FERTILIZING VALUE OF MAYON VOLCANO ASH: IV. EFFECTS UPON GROWTH AND DEVELOPMENT OF ABACÁ PLANTS [1]

VICENTE C. URETA

WITH FOUR TEXT FIGURES

The good growth of abacá plants in the vicinity of Mayon Volcano is believed to be partly due to the soil of volcanic origin.

To find out if Mayon Volcano ash has a favorable influence upon the growth of abacá, young plants were grown in Lipa clay loam in pots to which were added Mayon ash in varying amounts, with and without certain commercial fertilizers, such as ammonium sulfate and sodium nitrate.

Galvez (1939) found that Mayon volcanic ash contained SiO_2, TiO_2, Al_2O_3, Fe_2O_3, Mn_3O_4, CaO, MgO, K_2O, Na_2O, and P_2O_5. Galvez, Aquino, and Mamisao (1939) found that the volcanic ash contained less available K_2O but more available P_2O_5 than the Albay soils. They also found that the volcanic ash contained all the inorganic constituents generally present in ordinary agricultural soils except nitrogen and, with the exception of CaO, its fertilizing constituents, like K_2O and P_2O_5, were more abundant than those present in ordinary soils.

Espino (1939a) observed that Mayon Volcano ash when added to Lipa clay loam in liberal amounts seemed harmful to young rice, corn, and radish plants, but not to soybean. The plants were stunted in growth and had chlorotic leaves. Espino (1939b) observed, however, that the harmful effects of the volcanic ash were neutralized by the addition of ammonium sulfate to the soil, and concluded that the liberal addition of the volcanic ash to Lipa clay loam poor in nitrogen might have produced a physiological unbalanced solution in the culture medium.

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[1] Experiment Station contribution No. 1440. Prepared in the Department of Agricultural Botany under the direction of Professor R. B. Espino.

MATERIALS AND METHODS

Plants

The experiments were begun on April 12, 1939. Young abacá plants, variety Tañgoñgon, were used. The seeds were obtained from the Mampising Agricultural High School, Pantukan, Davao. They were soaked in warm water, 40°C., for 20 minutes (Ferrer and Espino, 1923) and were germinated in boxes. To stimulate the growth of the young plants, a little ammonium sulfate was added to the soil in the germinating boxes. When about two months old, the seedlings, which were eight centimeters high, were transplanted to the earthen pots.

Fig. 1.—Young abacá plants in pot cultures. Excepting I (soil only) each of the other cultures received 6.12 grams of ammonium sulfate and Mayon ash: III, 49 grams; IV, 98 grams; V, 196 grams, and VI, 392 grams. Grown from suckers, April 5 to August 23, 1940. Photograph by the Photographic Division, College of Agriculture.

Suckers from discontinued cultures were used in later experiments begun on April 5, 1940.

Soil

The Lipa clay loam which was used in this study was taken from a place behind the Administration Building of the College of Agriculture. The soil was dried in the air, sifted, and thoroughly mixed. Analysis of similar soil made by the Department of Agricultural Chemistry for Mr. José Madrigal Jr. showed that it contained in available form 0.00264 per cent N, 0.00279 per cent P_2O_5, and 0.02228 per cent K_2O. About thirty-five kilograms of the soil were placed in each culture pot.

Mayon ash

The Mayon ash was obtained from the same stock where Galvez (1939), Galvez, Aquino and Mamisao (1939), Espino (1939a, 1939b), and Espino and Agda (1941) secured the volcanic ashes they employed in their respective studies. The amount of Mayon ash needed was determined by first placing it on a concrete floor, then sifting it thoroughly, and weighing out the needed amounts. The Mayon ash was added to the soil before the seedlings or suckers of abacá were transplanted to the pots.

Culture media

Five sets of pot cultures were run from April, 1939 to October, 1940.

Fig. 2.—Young abacá plants in pot cultures. Excepting I (soil only) each of the other cultures received 12.3 grams of ammonium sulfate and Mayon ash: III, 49 grams; IV, 98; grams; V, 196 grams; and VI, 392 grams. Grown from suckers, April 5 to August 23, 1940. Photograph by the Photographic Division, College of Agriculture.

The first set consisted of seven quadruplicate cultures, six of which were supplied with varying amounts of Mayon ash, ranging from 12.25 grams in culture II to 392 grams (the highest amount tried) in culture VII. Culture I, also quadruplicate, received no Mayon ash and served as the control.

The second set consisted of six quadruplicate cultures: I contained only soil, II received 6.12 grams of ammonium sulfate per pot, and III to VI each had 6.12 grams of ammonium sulfate supplemented with varying amounts of Mayon ash, ranging from 49 grams in III to 392 grams, the highest, which was tried in VI.

The data in the second set and in all succeeding sets were obtained from the mother plant as well as from the suckers produced by it.

The third set consisted also of six quadruplicate cultures, which received treatments similar to those given in set 2 except that the ammonium sulfate was added to cultures II to VI at the rate of 12.25 grams per pot.

The fourth set had five quadruplicate cultures. Cultures II to V received 18.36 grams of ammonium sulfate each and Mayon ash ranging in quantity from 49 grams in II to 392 grams in V. The control cultures were included.

Fig. 3.—Young abacá plants in pot cultures. Excepting I (soil only) each of the other cultures received 18.4 grams of ammonium sulfate and Mayon ash: II, 49 grams; III, 98 grams; IV, 196 grams; and V, 392 grams. Grown from suckers, April 5 to August 23, 1940. Photograph by the Photographic Division, College of Agriculture.

The fifth set was composed of eight quadruplicate cultures. Cultures II to VIII received 6.12 grams of ammonium sulfate and 3.06 grams of sodium nitrate each. In cultures III to VIII, varying amounts of Mayon ash were added, ranging from 12.25 grams in III to 392 grams in VIII. Unfertilized cultures were also included as control.

Care of cultures

The cultures were grown in the open and exposed to the rain and sunshine. They were watered almost every morning and afternoon. Weeding and cultivation were done whenever needed.

EXPERIMENTS AND RESULTS

Mayon Volcano ash in varying amounts

Mayon Volcano ash was the first set of cultures tried. The abacá seedlings were transplanted to the pots on February 6, 1939; the Mayon ash was added on April 12, 1939; and the experiment was terminated on April 3, 1940. The heights of the false stems were measured weekly in order to determine the comparative effects of the amounts of Mayon ash. Similarly, once a week during the duration of this experiment, the leaves in the different cultures were counted. From these measurements and observations, data on weekly growth of false stems and on weekly production of leaves were computed.[2]

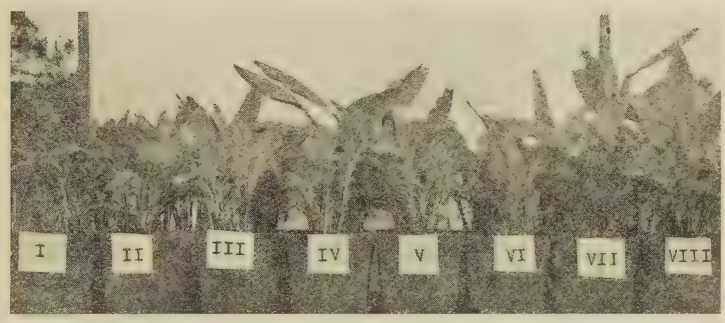

Fig. 4.—Young abacá plants in pot cultures. Excepting I (soil only) each of the other cultures received 6.12 grams of ammonium sulfate and 3.06 grams of sodium nitrate to which was added Mayon ash: III, 12.3 grams; IV, 24.5 grams; V, 49 grams; VI, 98 grams; VII, 196 grams; and VIII, 392 grams. Grown from suckers, April 5 to August 23, 1940. Photograph by the Photographic Division, College of Agriculture.

Mixtures of Mayon ash and ammonium sulfate

The three sets of cultures supplied with mixtures of Mayon ash and ammonium sulfate were simultaneously started on April 5, 1940. Instead of seedlings, young suckers of abacá obtained from the preceding cultures were planted in the pots. The photographs of the cultures which were taken before they were harvested are shown in figures 1 to 3. The experiment was concluded on August 23, 1940.

[2] The data are in the files of the Department of Agricultural Botany.

Experimental data from the quadruplicate cultures were gathered. Only averages with the corresponding standard errors are shown in tables 1a, b, c. Data on weekly growth of the false stems as well as on weekly production of leaves were also gathered [3].

Mixtures of Mayon ash, ammonium sulfate, and sodium nitrate

This set was run simultaneously with the preceding three sets of cultures. It was started on April 5, 1940 and concluded on August 23, 1940. The cultures were also photographed (fig. 4), and the experimental data gathered. Only the averages with their corresponding standard errors are recorded in table 1d. The average data on weekly growth increments of the false stems and those of the weekly production of leaves were also gathered [4].

Criteria of results

Weekly growth of false stem. The height of a false trunk was measured weekly from the lid of the pot as a base mark to the point of intersection of the petioles of the two youngest leaves. The weekly growth increments of four similar cultures were averaged.

Number of leaves. Once a week, the leaves of the plants in each pot were counted. The results from four similar cultures were averaged.

Leaf product. The leaf product was obtained by multiplying the length by the width at the widest portion of the blade of each leaf. The data from similar cultures were averaged.

Fresh weight of top. The false trunk was cut close to the ground level, and the top was immediately weighed. The weights of tops (false stem and leaves) from similar cultures were averaged.

Length and number of roots. The roots were dug up from each pot, washed with water, and counted. The lengths of the different roots were measured separately, and the results obtained were averaged.

Fresh weight of roots. The roots were weighed while fresh and the results from the quadruplicate cultures were averaged.

[3, 4] See footnote 2.

DISCUSSION OF RESULTS

Effects of Mayon ash on weekly growth of false stem

In order to facilitate the comparison and selection of cultures on the basis of weekly growth, the three highest growth increments were arbitrarily selected each week. In table A are given the selected growth data and the total growths of false stems from the different cultures.

TABLE A

CULTURE NO.	DURING FIFTY-ONE WEEKS			SELECTED DATA	
	Amount of Mayon ash used	Total av. growth of false stems	Total av. number of leaves produced	Number of leaves	On weekly growth
	grams	*cm.*			
I	0	23.65	32.00	22	15
II	12.5	30.85	34.08	20	31
III	24.5	30.44	28.75	14	32
IV	49.0	25.44	33.25	17	19
V	98.0	23.74	31.85	19	16
VI	196.0	24.96	29.00	12	19
VII	392.0	26.30	37.10	18	13

As shown in table A, the growths of the abacá plants or of their false stems did not go with or against the amounts of Mayon ash used. Compared with the control, however, the Mayon ash-treated cultures grew generally a little faster, and thus indicated that the Mayon ash when added to Lipa clay loam was not poisonous, and it was probably even beneficial to the abacá plants.

To locate the fast growing cultures and to know the growth reactions of the plants to the treatments given, the three highest growth records each week were arbitrarily selected. The results show that the selected growth records are more or less equally distributed among the cultures that received ammonium sulfate and varying amounts of Mayon ash. It was observed that applications of mixtures of Mayon ash and ammonium sulfate were apparently more conducive to rapid growth of false stems of the young abacá plants than the control, and beneficial effects were derived from the ammonium sulfate as well as from the Mayon ash.

The total number of these selected data on weekly growth under each culture was determined, and the results are recorded in table B.

TABLE B

CULTURE NO.	MAYON ASH	NUMBER OF SELECTED DATA ON WEEKLY GROWTH			
		6.12 gr. ammonium sulfate	12.25 gr. ammonium sulfate	18.36 gr. ammonium sulfate	6.12 gr. ammonium sulfate 3.06 gr. sodium nitrate
	grams				
I	0	1	0	1	1
II	0	5	3	blank	6
III	49	12	12	12	4
IV	98	8	15	15	5
V	196	18	18	18	10
VI	392	10	12	15	11

Table B shows that cultures III to VI each received more of the selected growth records than the control, or culture II, which was supplied only with ammonium sulfate. It is again apparent that the Mayon ash was beneficial to the abacá plants, especially when supplemented with ammonium sulfate.

Effects of Mayon ash on weekly production of abacá leaves

The data on hand show that varying the amounts of Mayon ash added to Lipa clay loam seems not to have influenced the production of leaves one way or another, for the arbitrarily selected cultures are scattered all over the table, irrespective of the amount of Mayon ash used, and the control was not always the slowest in the production of leaves. Moreover, the total number of leaves produced and the selected data on leaf production from the different cultures (table A) show high values both from the control and some of the Mayon ash-treated cultures. Thus, it appears that in the production of abacá leaves, an addition of Mayon ash to Lipa clay loam, a soil poor in N, fairly rich in K_2O, and with a fair content of P_2O_5, was not beneficial.

In order that the fast leaf-producing cultures and the reactions of the abacá plants to the different treatments could be located, the three highest leaf production records were arbitrarily selected each week. The results show that the selected data are scattered all over the tables opposite the cultures that were supplied with ammonium sulfate or ammonium sulfate and sodium nitrate and varying amounts of Mayon ash. And, to show easier comparisons of the different cultures, especially in connection with effects on leaf production by

varying the amount of ammonium sulfate and of Mayon ash, table C is presented here.

TABLE C

CULTURE NO.	MAYON ASH	NUMBER OF SELECTED DATA ON NUMBER OF LEAVES			
		6.12 gr. ammonium sulfate	12.25 gr. ammonium sulfate	18.36 gr. ammonium sulfate	6.12 gr. ammonium sulfate 3.06 gr. sodium nitrate
	grams				
I	0	4	1	2	0
II	0	5	7	blank	5
III	49	11	9	10	7
IV	98	13	12	16	10
V	196	14	17	13	6
VI	392	12	13	15	11

Table C shows that culture II is better than I, and I is worse than any of the other cultures. Moreover, any of the cultures (III to VI) which received varying amounts of Mayon ash in addition to the amount of nitrogenous fertilizer or fertilizers is better than II, which received only the nitrogenous fertilizers. Therefore, it appears that the addition of Mayon ash when accompanied by ammonium sulfate alone or with sodium nitrate improved the production of leaves of young abacá plants.

Effects upon abacá plants of mixture of ammonium sulfate and Mayon ash

Tables 1a, b, c show that under the seven criteria of results employed, culture I, in general, is worse than any of the other cultures, and culture II in table 1a or 1b produced harvest data lower than those obtained from any of the cultures from III to VI. The addition of Mayon ash to Lipa clay loam when accompanied with ammonium sulfate seems to have improved the growth and development of the young abacá plants.

The cultures in each of the tables 1a, b, c were statistically compared with one another to verify the correctness of these casual observations. The results (tables 2a, b, c) show that culture I, which contained only the soil, was significantly inferior to any of the cultures that received ammonium sulfate alone or was supplemented with Mayon ash.

Culture II, which received only ammonium sulfate amounting to 6.72 or 12.25 grams per pot (the 18.36 grams were inadvertently not tried), produced harvest data under the criteria of weight of tops, weight of roots, and leaf products always lower than the corresponding harvest data obtained from any of the cultures from III to VI (table 1a). In number of roots and suckers, however, the differences are mostly statistically insignificant (table 2a). In spite of these discrepancies, it may still be concluded that the addition of Mayon ash to Lipa clay loam supplemented with 8.12 or 12.25 grams of ammonium sulfate per pot increased the fresh weights of tops and of roots as well as the area (leaf-product) of leaves of abacá plants.

The most promising cultures were next determined. The two highest harvest data under each criterion of results are arbitrarily selected and underlined in tables 1a, b. c. The results show that most of the underlined data belong to cultures V and VI. For further comparison, the selected cultures are transferred to table D.

TABLE D

CULTURE NO.	TREATMENT		HEIGHT OF PLANTS	FRESH WEIGHT		NUMBER OF ROOTS	LEAF PRODUCT	NUMBER OF SUCKERS
	Mayon ash	Ammonium sulfate		Tops	Roots			
	grams	*grams*	*cm.*	*grams*	*grams*		*sq. cm.*	
V table 1a	196	6.12	129	965	327	114	7512	*6.25*
VI table 1a	392	6.12	103	1579	402	153	10253	5.25
V table 1b	196	12.25	*145*	1955	*815*	*221*	10683	*5.50*
VI table 1b	392	12.25	112	*2056*	635	*182*	*12312*	4.75
IV table 1c	196	18.36	*143*	1918	737	139	10348	4.50
V table 1c	392	18.36	123	*2302*	*962*	162	*13902*	3.75

Most of the underlined data in table D fall in cultures V and VI in table 1b. These cultures may be considered the most promising in the lots. As shown in table 2b, cultures V and VI are statistically superior to any of the ammonium sulfate plus Mayon ash treated cultures in the same table. But under four criteria, V is better than VI, whereas under three other criteria, VI is superior to V. In table 1b or table 2b, these two cultures are the best. Both received the

same amount of ammonium sulfate, but V was further treated with 196 grams of Mayon ash, and VI, with 392 grams of the ash. Since the selected cultures had 196 or 392 grams of Mayon ash (table D), and since fairly high harvest data are also found in cultures IV and V from table 1c, might be safe to conclude that the most promising treatment for the young abacá plant when grown in Lipa clay loam should be an application of 196 to 392 grams of Mayon ash supplemented with 12.25 to 18.36 grams of ammonium sulfate.

Effects upon abacá plants of mixtures of ammonium sulfate, sodium nitrate, and Mayon ash

In this set of cultures, I, the control, again produced harvest data statistically lower than the corresponding data from the fertilized cultures. Moreover, like the previous sets of cultures (those supplied with N only from ammonium sulfate), culture II, which received ammonium sulfate and sodium nitrate but no Mayon ash, produced harvest data mostly statistically poorer than the corresponding harvest data obtained from any of the fertilized cultures at the same time treated with Mayon ash. The addition of Mayon ash to Lipa clay loam supplemented with ammonium sulfate and sodium nitrate, therefore, considerably improved the growth and development of the young abacá plants. Although the development of the plants was improved by the application of the two nitrogenous fertilizers, the addition of Mayon ash in itself brought about a little further improvement in the growth and development of young abacá plants.

Examination of the data in tables 1a, 1b, 1c, and 1d reveals two things; first, that the amounts of N in the two promising cultures in tables 1a, 1b, and 1c are more than those in any of the cultures in table 1d, and second, most of the harvest data in the first three tables are greater than the corresponding harvest data contained in table 1d. Therefore, owing to the apparent differences in nitrogen contents, it seems best not to make further comparisons.

It should, however, be pointed out that in the production of suckers, a mixture of Mayon ash, ammonium sulfate, and sodium nitrate was more effective than a mixture of Mayon ash and ammonium sulfate (tables 1a, b, c, d).

SUMMARY

1. The applications of mixtures of Mayon ash and ammonium sulfate were probably more conducive to rapid growth of false stems of the young abacá plants than the control, and the beneficial effects were derived from the ammonium sulfate as well as from the Mayon ash.

2. In the production of abacá leaves, the addition of Mayon ash to Lipa clay loam, a soil poor in N, fairly rich in K_2O, and with a fair content of P_2O_5, was not beneficial.

3. The addition of Mayon ash when accompanied by ammonium sulfate alone or with sodium nitrate apparently improved the production of leaves of young abacá plants.

4. The addition of Mayon ash to Lipa clay loam when accompanied with ammonium sulfate seems to have improved the growth and development of the young abacá plants.

5. The addition of Mayon ash to Lipa clay loam supplemented with 6.12 or 12.25 grams of ammonium sulfate per pot increased the weight of tops and of roots as well as the area (leaf-product) of the leaves of abacá plants.

6. The best or most promising treatment for the young abacá plant when grown in Lipa clay loam should be an application of 196 to 392 grams of Mayon ash accompanied by or supplemented with 12.3 to 18.4 grams of ammonium sulfate.

7. The addition of Mayon ash to Lipa clay loam supplemented with ammonium sulfate and sodium nitrate considerably improved the growth and development of the young abacá plants.

8. Although the development of the plants was improved by the application of the two nitrogenous fertilizers, the addition of Mayon ash in itself brought about a little further improvement in the growth and development of young abacá plants.

9. In the production of suckers, a mixture of Mayon ash, ammonium sulfate, and sodium nitrate seems to be more effective than a mixture of Mayon ash and ammonium sulfate.

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TABLE 1a

Average data obtained from young abacá plants grown in Lipa clay loam supplied with varying amounts of Mayon Volcano ash and 6.12 grams of ammonium sulfate [a]

CULTURE NUMBER	TREATMENTS		HEIGHT	FRESH WEIGHT OF TOPS	FRESH WEIGHT OF ROOTS	LENGTH OF ROOTS	NUMBER OF ROOTS	LEAF PRODUCT	SUCKERS PRODUCED
	Mayon ash	Ammonium sulfate							
	grams	*grams*	*cm.*	*grams*	*grams*	*cm.*		*sq. cm.*	
I	0	0	28.25	356.0∓40.57	55.3∓7.24	93.0∓17.16	35.8∓7.05	3789.2∓119.3	3.5∓0.75
II	0	6.12	59.35	590.2∓28.79	222.0∓14.16	109.2∓22.57	97.8∓8.15	4156.3∓219.5	*5.8*∓0.45
III	49	6.12	77.52	768.2∓17.01	308.5∓29.84	*159.8*∓10.00	101.5∓1.98	4999.3∓102.3	4.5∓0.41
IV	98	6.12	78.25	812.2∓36.06	316.2∓15.06	104.8∓6.51	110.3∓10.08	6642.2∓242.1	5.3∓0.64
V	196	6.12	*128.95*	*964.5*∓36.34	*327.0*∓25.75	*116.5*∓5.42	*113.5*∓15.14	*7511.8*∓321.6	*6.3*∓1.13
VI	392	6.12	*103.00*	*1579.2*∓28.74	*401.5*∓11.48	107.8∓21.02	*153.2*∓5.50	*10252.6*∓169.7	5.3∓0.57

[a] Experiment from April 5 to August 23, 1940.

TABLE 1b

Average data obtained from young abacá plants grown in Lipa clay loam supplied with varying amounts of Mayon Volcano ash and 12.25 grams of ammonium sulfate [a]

CULTURE NUMBER	TREATMENTS		HEIGHT	FRESH WEIGHT OF TOPS	FRESH WEIGHT OF ROOTS	LENGTH OF ROOTS	NUMBER OF ROOTS	LEAF PRODUCT	SUCKERS PRODUCED
	Mayon ash	Ammonium sulfate							
	grams	*grams*	*cm.*	*grams*	*grams*	*cm.*		*sq. cm.*	
I	0	0	28.25	256.0∓40.51	55.3∓7.24	93.0∓17.16	35.8∓7.05	3769.2∓113.3	3.5∓0.75
II	0	12.25	68.05	867.7∓22.16	247.8∓20.90	*137.8*∓7.12	74.3∓13.15	5918.3∓562.0	4.8∓0.96
III	49	12.25	108.30	1239.5∓30.76	407.3∓16.98	105.5∓0.95	122.8∓12.51	8210.7∓108.5	*5.5*∓0.55
IV	98	12.25	*113.40*	1388.2∓43.48	504.5∓13.33	120.8∓10.72	127.3∓10.55	9959.0∓413.9	5.0∓0.93
V	196	12.25	*144.67*	*1954.5*∓56.01	*815.3*∓32.07	*127.0*∓22.32	*220.3*∓17.02	*10683.5*∓258.2	*5.5*∓0.75
VI	392	12.25	112.25	*2080.5*∓27.69	*634.8*∓28.43	93.5∓4.75	*181.5*∓8.10	*12317.4*∓113.3	4.8∓0.96

[a] Experiment from April 5 to August 23, 1940.

TABLE 1c

Average data obtained from young abacá plants grown in Lipa clay loam supplied with varying amounts of Mayon Volcano ash and 18.36 grams of ammonium sulfate [a]

CULTURE NUMBER	TREATMENTS		HEIGHT	FRESH WEIGHT OF TOPS	FRESH WEIGHT OF ROOTS	LENGTH OF ROOTS	NUMBER OF ROOTS	LEAF PRODUCT	SUCKERS PRODUCED
	Mayon ash	Ammonium sulfate							
	grams	*grams*	*cm.*	*grams*	*grams*	*cm.*		*sq. cm.*	
I	0	0	28.25	356.0∓40.51	55.3∓7.24	93.0∓12.16	35.8∓7.05	3769.2∓1.35	3.5∓0.75
II	49	18.36	117.82	1422.2∓27.49	530.3∓38.30	108.0∓5.35	127.3∓13.61	4999.4∓101.66	*5.3*∓8.92
III	98	18.36	*136.53*	1863.0∓8.05	693.8∓7.25	*115.8*∓5.16	133.5∓9.68	*11522.9*∓108.25	*4.5*∓0.95
IV	196	18.36	*142.77*	*1918.2*∓22.43	*737.3*∓45.32	*113.5*∓5.14	*139.3*∓9.98	10348.0∓348.00	4.5∓1.17
V	392	18.36	122.67	*2301.5*∓26.47	*962.0*∓21.11	108.0∓1.60	*162.0*∓21.74	*13902.8*∓178 39	3.8∓0.54

[a] Experiment, from April 5 to August 23, 1940.

TABLE 1d

Average data obtained from young abacá plants grown in Lipa clay loam supplied with varying amounts of Mayon Volcano ash and the same amounts of ammonium sulfate and sodium nitrate [a]

CULTURE NUMBER	TREATMENTS			HEIGHT	FRESH WEIGHT OF TOPS	FRESH WEIGHT OF ROOTS	LENGTH OF ROOTS	NUMBER OF ROOTS	LEAF PRODUCT	SUCKERS PRODUCED
	Mayon ash	Ammonium sulfate	Sodium nitrate							
	grams	*grams*	*grams*	*cm.*	*grams*	*grams*	*cm.*		*sq. cm.*	
I	0	0	0	28.25	356.0∓40.47	55.3∓7.24	93.0∓17.16	35.8∓7.05	3189.2∓113.3	3.5∓0.75
II	0	6.12	3.06	99.48	621.2∓32.47	238.2∓17.54	103.5∓12.50	89.0∓5.89	4844.9∓258.8	6.8∓0.54
III	12.25	6.12	3.06	*112.75*	889.7∓22.11	343.7∓15.05	102.2∓17.15	108.7∓8.03	6511.8∓356.3	6.3∓0.89
IV	24.50	6.12	3.06	*128.43*	*899.2*∓22.21	*418.7*∓25.20	93.5∓17.16	122.2∓18.68	*8894.5*∓441.4	*8.3*∓1.24
V	49.00	6.12	3.06	98.70	646.2∓32.24	*405.5*∓10.07	102.0∓29.33	119.7∓16.20	5917.1∓725.5	6.5∓1.08
VI	98.00	6.12	3.06	103.43	797.5∓12.21	332.2∓18.29	*146.5*∓26.73	*123.2*∓5.82	7168.3∓113.3	*7.8*∓1.63
VII	196.00	6.12	3.06	108.65	837.7∓40.57	373.7∓17.24	109.7∓9.90	*123.2*∓11.04	*8389.6*∓130.8	4.8∓0.54
VIII	392.00	6.12	3.06	111.23	*1058.0*∓32.32	305.0∓21.79	*110.7*∓17.40	*124.2*∓7.50	7253.3∓228.6	4.0∓0.35

[a] Experiment from April 5 to August 23, 1940.

TABLE 2a

Statistical comparisons of average data obtained from young abacá plants grown in Lipa clay loam supplied with varying amounts of Mayon ash and 6.12 grams of ammonium sulfate

CULTURES COMPARED	FRESH WEIGHT OF TOPS [a]	FRESH WEIGHT OF ROOTS [a]	NUMBER OF ROOTS [a]	LEAF PRODUCTS [a]	NUMBER OF SUCKERS [a]
	grams	*grams*		*sq. cm.*	
II–I	234.2 ∓ 49.7 (S)	166.8 ∓ 15.9 (S)	62.0 ∓ 10.8 (S)	387.1 ∓ 247.0 (I)	2.25 ∓ 0.87 (S)
III–I	412.2 ∓ 44.0 (S)	253.3 ∓ 30.7 (S)	65.8 ∓ 7.3 (S)	1230.1 ∓ 152.6 (S)	1.00 ∓ 0.85 (I)
IV–I	456.2 ∓ 54.3 (S)	261.0 ∓ 16.7 (S)	74.5 ∓ 12.3 (S)	3872.9 ∓ 267.3 (S)	1.75 ∓ 0.99 (I)
V–I	608.5 ∓ 54.3 (S)	271.8 ∓ 26.7 (S)	77.8 ∓ 8.9 (S)	3742.6 ∓ 248.8 (S)	2.75 ∓ 1.36 (S)
VI–I	1223.2 ∓ 49.7 (S)	346.3 ∓ 13.6 (S)	117.5 ∓ 16.7 (S)	6483.3 ∓ 204.0 (S)	1.75 ∓ 0.94 (I)
III–II	178.0 ∓ 33.4 (S)	86.5 ∓ 33.0 (S)	3.8 ∓ 8.4 (I)	843.1 ∓ 242.2 (S)	—1.50 ∓ 0.61 (S)
IV–II	222.0 ∓ 46.1 (S)	94.2 ∓ 20.7 (S)	12.5 ∓ 13.0 (I)	2485.9 ∓ 326.7 (S)	—0.50 ∓ 0.79 (I)
V–II	374.3 ∓ 46.4 (S)	105.0 ∓ 29.4 (S)	15.8 ∓ 9.8 (I)	3355.6 ∓ 311.9 (S)	0.50 ∓ 1.22 (I)
VI–II	989.0 ∓ 40.7 (S)	179.5 ∓ 18.2 (S)	55.3 ∓ 17.2 (S)	6098.4 ∓ 27.7 (S)	—0.50 ∓ 0.72 (I)
IV–III	44.0 ∓ 39.9 (I)	7.7 ∓ 33.4 (I)	8.8 ∓ 10.3 (I)	1642.8 ∓ 262.8 (S)	1.00 ∓ 0.72 (I)
V–III	196.3 ∓ 40.1 (S)	18.5 ∓ 39.4 (I)	12.0 ∓ 5.8 (S)	2512.5 ∓ 244.0 (S)	2.00 ∓ 1.21 (I)
VI–III	811.0 ∓ 33.4 (S)	93.0 ∓ 32.0 (S)	51.7 ∓ 13.3 (S)	5253.0 ∓ 198.1 (S)	1.00 ∓ 0.70 (I)
V–IV	152.3 ∓ 51.2 (S)	10.8 ∓ 29.8 (I)	3.3 ∓ 11.5 (I)	869.7 ∓ 328.2 (S)	1.00 ∓ 1.31 (I)
VI–IV	762.0 ∓ 46.1 (S)	85.3 ∓ 19.0 (S)	43.0 ∓ 18.9 (S)	3611.5 ∓ 295.6 (S)	0.00 ∓ 0.86 (I)
VI–V	645.9 ∓ 46.3 (S)	74.5 ∓ 28.2 (S)	39.7 ∓ 16.1 (S)	2740.8 ∓ 279.1 (S)	1.00 ∓ 1.27 (I)

[a] (S) means significant and (I) insignificant.

TABLE 2b

Statistical comparisons of average data obtained from young abacá plants grown in Lipa clay loam supplied with varying amounts of Mayon ash and 12.25 grams of ammonium sulfate

CULTURES COMPARED	FRESH WEIGHT OF TOPS [a]	FRESH WEIGHT OF ROOTS [a]	NUMBER OF ROOTS [a]	LEAF PRODUCTS [a]	NUMBER OF SUCKERS [a]
	grams	*grams*		*sq. cm.*	
II–I	511.7 ∓ 46.2 (S)	192.5 ∓ 69.6 (S)	38.5 ∓ 14.9 (S)	2149.1 ∓ 573.3 (S)	1.3 ∓ 1.2 (I)
III–I	883.5 ∓ 50.9 (S)	352.0 ∓ 14.0 (S)	87.0 ∓ 14.4 (S)	4441.5 ∓ 156.8 (S)	2.0 ∓ 0.9 (S)
IV–I	1032.2 ∓ 42.8 (S)	449.3 ∓ 15.2 (S)	81.5 ∓ 12.7 (S)	6190.6 ∓ 429.1 (S)	1.5 ∓ 1.2 (I)
V–I	1598.5 ∓ 69.2 (S)	760.0 ∓ 32.8 (S)	185.0 ∓ 18.4 (S)	6914.3 ∓ 281.8 (S)	2.0 ∓ 1.0 (I)
VI–I	1724.5 ∓ 49.1 (S)	579.5 ∓ 29.5 (S)	145.8 ∓ 10.7 (S)	8548.2 ∓ 441.0 (S)	1.3 ∓ 0.9 (I)
III–II	371.8 ∓ 37.9 (S)	159.0 ∓ 24.1 (S)	48.5 ∓ 18.5 (S)	2292.4 ∓ 572.3 (S)	0.8 ∓ 1.1 (I)
IV–II	520.5 ∓ 48.8 (S)	256.8 ∓ 24.8 (S)	53.0 ∓ 16.9 (S)	4041.6 ∓ 697.9 (S)	0.3 ∓ 1.3 (I)
V–II	1086.8 ∓ 60.2 (S)	567.5 ∓ 28.3 (S)	146.0 ∓ 21.5 (S)	4765.2 ∓ 618.4 (S)	0.8 ∓ 1.2 (I)
VI–II	1212.8 ∓ 35.5 (S)	387.0 ∓ 35.3 (S)	107.3 ∓ 15.4 (S)	6399.2 ∓ 705.4 (S)	0.0 ∓ 1.2 (I)
IV–III	148.7 ∓ 52.3 (S)	97.3 ∓ 17.9 (S)	4.5 ∓ 16.4 (I)	1749.1 ∓ 427.8 (S)	−0.5 ∓ 1.1 (I)
V–III	715.0 ∓ 63.9 (S)	408.0 ∓ 34.3 (S)	98.0 ∓ 21.5 (S)	2472.8 ∓ 279.0 (S)	0.0 ∓ 0.9 (I)
VI–III	841.0 ∓ 41.4 (S)	227.5 ∓ 30.9 (S)	58.8 ∓ 15.3 (S)	4106.7 ∓ 439.8 (S)	−0.8 ∓ 0.7 (I)
V–IV	466.3 ∓ 70.9 (S)	310.8 ∓ 34.7 (S)	93.5 ∓ 20.0 (S)	723.7 ∓ 487.8 (S)	0.5 ∓ 1.2 (I)
VI–IV	692.3 ∓ 51.4 (S)	134.1 ∓ 31.4 (S)	54.3 ∓ 13.2 (S)	2357.6 ∓ 594.2 (S)	−0.3 ∓ 1.1 (I)
VI–V	126.0 ∓ 62.5 (S)	−176.5 ∓ 42.9 (S)	−39.3 ∓ 18.8 (S)	1633.9 ∓ 498.3 (S)	−0.8 ∓ 0.9 (I)

[a] (S) means significant and (I) insignificant.

TABLE 2c

Statistical comparisons of average data obtained from young abacá plants grown in Lipa clay loam supplied with varying amounts of Mayon ash and 18.36 grams of ammonium sulfate

CULTURES COMPARED	FRESH WEIGHT OF TOPS [a]	FRESH WEIGHT OF ROOTS [a]	NUMBER OF ROOTS [a]	LEAF PRODUCTS [a]	NUMBER OF SUCKERS [a]
	grams	*grams*		*sq. cm.*	
II–I	1066.2 ∓ 49.2 (S)	475.0 ∓ 39.0 (S)	91.5 ∓ 15.3 (S)	1230.18 ∓ 152.1 (S)	1.8 ∓ 1.2 (I)
III–I	1507.0 ∓ 41.4 (S)	638.5 ∓ 19.4 (S)	92.5 ∓ 12.0 (S)	7753.7 ∓ 156.5 (S)	1.0 ∓ 1.2 (I)
IV–I	1562.2 ∓ 46.4 (S)	682.0 ∓ 45.9 (S)	103.5 ∓ 12.2 (S)	7578.8 ∓ 559.6 (S)	1.0 ∓ 1.4 (I)
V–I	1945.5 ∓ 48.4 (S)	906.8 ∓ 22.3 (S)	126.3 ∓ 22.9 (S)	10135.6 ∓ 212.0 (S)	0.3 ∓ 0.9 (I)
III–II	440.8 ∓ 29.0 (S)	163.5 ∓ 42.3 (S)	6.3 ∓ 16.7 (I)	6523.5 ∓ 148.2 (S)	−0.8 ∓ 1.5 (I)
IV–II	496.0 ∓ 35.8 (S)	207.0 ∓ 59.4 (S)	12.0 ∓ 16.9 (I)	537.4 ∓ 557.4 (I)	−0.8 ∓ 1.5 (I)
V–II	1079.3 ∓ 38.5 (S)	431.8 ∓ 43.8 (S)	34.8 ∓ 25.6 (I)	8903.5 ∓ 205.6 (S)	−1.5 ∓ 1.0 (I)
IV–III	55.2 ∓ 23.8 (S)	43.5 ∓ 48.7 (I)	5.8 ∓ 13.9 (I)	−1174.9 ∓ 558.5 (S)	0.0 ∓ 1.5 (I)
V–III	438.5 ∓ 27.7 (S)	226.3 ∓ 27.8 (S)	28.5 ∓ 23.8 (I)	2379.9 ∓ 208.9 (S)	−0.7 ∓ 1.1 (I)
V–IV	383.3 ∓ 34.7 (S)	182.2 ∓ 50.0 (S)	27.8 ∓ 23.9 (I)	3554.8 ∓ 576.4 (S)	−0.7 ∓ 1.3 (I)

[a] (S) means significant and (I) insignificant.

COMPARATIVE EFFECTS OF FOUR COMMERCIAL FERTILIZERS UPON THE YIELD OF NATIVE YELLOW FLINT CORN [1]

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Studies on the effects of the application of commercial fertilizers on the yield of corn have been conducted in the College of Agriculture. Inasmuch as, no study has been made of the comparative effects of the same amount of different commercial fertilizers, upon the yield of corn, an experiment was conducted by the writer from May 15 to August 30, 1940.

Vibar (1911) found that basic slag, tankage, and kainit when applied singly or in combination of any two increased the yield of corn, and the application of the three in combination produced the highest yield.

Montellano (1916) found that a phosphatic fertilizer in the form of superphosphate increased the yield of corn. When phosphoric acid and nitrogen were combined, however, the increase was greater. Potash in combination with phosphoric acid also increased the yield, but nitrogen, phosphoric acid, and potash (N-P-K) in combination obtained the greatest yield.

Villa [2] made a study on different amounts of Nitrophoska IG No. 2 (N-14, P-11.7, K-26) applied to corn. He found that the 100-kilogram application per hectare gave the highest yields of corn for the two seasons.

Yatar [3] found, among other amounts used in his experiment on corn with Leunaphos IG No. 2 (N-16.5, P-20, K-0), that the 100-

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[1] Experiment Station contribution No. 1443. Read before the Los Baños Biological Club on June 26, 1941.

[2] VILLA, ISIDRO C. The effects upon the yield of corn of the application of different rates of a fertilizer containing 14 per cent nitrogen, 11.7 per cent phosphoric acid and 26 per cent potash (Nitrophoska IG No. 2). (Thesis presented for graduation with the degree of Bachelor of Science in Agriculture from the College of Agriculture. 1934. Unpublished).

[3] YATAR, PEDRO. The effects of different rates of application of Leunaphos fertilizer on corn. (Thesis presented for graduation with the degree of Bachelor of Science in Agriculture from the College of Agriculture. 1934. Unpublished).

kilogram application per hectare gave the highest yields of corn for the two seasons of culture.

Braga [4] in his work on the effects of the application of different amounts of superphosphate (N-0, P-18-20, K-0) upon the yield of corn found that the 100-kilogram rate per hectare gave the highest average yield of grain and stover in both seasons.

Crisologo [5] in his experiment on the effects upon the yield of corn of the application of different amounts of ammonium phosphate (N-20, P-20, K-0), found that 100 kilograms per hectare applied to corn plants yielded the most in both the dry and wet season cultures.

MATERIALS AND METHODS

Corn variety and seed used

The corn used in this study was the Native Yellow Flint. The seeds were obtained from the crop raised in 1939 on the Rural High School farm.

Plots and preparation

A field measuring 5,000 square meters was used in this experiment. It was divided into five equal blocks, each of which was again divided into five plots of ten by twenty meters. The particular treatment given to each plot was made at random.

The land was plowed three times at an interval of about one week between plowings and harrowed once after each plowing.

Planting the corn crop

Furrows were made about one meter apart with an animal-drawn plow. The corn seeds were planted by hand on May 29, 1940, in hills 80 centimeters apart in the row. Four or five seeds were dropped in each hill and covered with soil by the feet.

Cultivating, weeding, and thinning. The plants were cultivated twice with a plow for the first time when they were about three weeks old; and for the second, about one week later. Soon after the second

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[4] BRAGA, ADRIANO S. The effects upon the yield of corn of the application of differents amount of superphosphate. (Thesis presented for graduation with the degree of Bachelor of Science in Agriculture from the College of Agriculture. 1938. Unpublished).

[5] CRISOLOGO, JESUS MA. The effects upon the yield of corn of the application of different amounts of ammonium phosphate fertilizer containing 20 per cent nitrogen 20 per cent phosphoric acid (Amo-phos). (Thesis presented for graduation with the degree of Bachelor of Science in Agriculture from the College of Agriculture. 1938. Unpublished).

cultivation, the plants were thinned to three per hill. During the growing period, weeds were hoed down from time to time. All barren corn plants were removed before flowering.

Fertilizers and application

The fertilizers used were Superphosphate, Leunaphos IG No. 1, Leunaphos IG No. 2, and Nitrophoska IG No. 3. They were bought from Menzi and Company, Manila. According to the manufacturer, Superphosphate contains 18-20 per cent phosphoric acid; Leunaphos IG No. 1, 19.5 per cent nitrogen and 20 per cent phosphoric acid; Leunaphos IG No. 2, 16.5 per cent nitrogen and 20 per cent phosphoric acid; and Nitrophoska IG No. 3, 15 per cent nitrogen, 15 per cent phosphoric acid, and 18 per cent potash by analysis.

The fertilizers in powder form were applied separately and as uniformly as possible around the base of the plants at a radius of 10 to 15 centimeters. Eight grams, or 100 kilograms per hectare were applied to each hill. A bamboo tube with a capacity of about eight grams was used to facilitate the application. The fertilizers were applied about four weeks after planting or one day before the second cultivation that they might be covered as the plow was passed between the rows.

Harvesting, drying, and shelling. The ears were harvested on August 29, 1940 by snapping and husking them from the standing stalks. The harvest from each plot was dried separately. The ears were shelled with a corn sheller and the grains were weighed.

RESULTS AND DISCUSSION

Field observation

The corn seeds germinated in three to four days and the plants flowered in five to six weeks. The corn plants matured in about 100 days after planting. In general, the plants in the fertilized plots were darker green and more vigorous than those in the control. No marked difference occurred in the color of the leaves and stand of the plants in the different fertilized plots.

Yield

Table 1 shows the actual yields of the different plots. The highest yield, 59.28 kilograms, was obtained from Nitrophoska IG No. 3 in block 4. The same fertilizer in block 1 produced 59.00 kilo-

grams, a very close second. Control in block 1 gave the lowest yield, 26.72 kilograms.

This table shows that Nitrophoska IG No. 3 also gave the highest mean yield (53.604 kilograms), followed by Leunaphos IG No. 2 (48.418 kilograms), Leunaphos IG No. 1 (46.604 kilograms), and Superphosphate (44.496 kilograms). The control gave the lowest, 36.654 kilograms.

Table 2 shows the analysis of variance for yields in the blocks. It will be noticed that the variance owing to blocks was not significant and thus indicated that the variability in blocks did not affect the experiment. The fertility of the soil might be the same throughout, as the differences in yields of the different plots are not due to block assignment. The variance due to treatment was highly significant, as the estimated variance exceeds the value of F at one per cent level of significance. The different treatments were responsible for the difference in yields obtained.

A comparison of mean yield of the different plots is shown in table 3. Based on the mean yield of the control, the greatest increase was obtained from the plots fertilized with Nitrophoska IG No. 3, followed by Leunaphos IG No. 2, and Leunaphos IG No. 1. The least increase was from plots fertilized with Superphosphate.

The difference in yield for Superphosphate and the control is not significant. The value of the difference for Leunaphos IG No. 1 (9.950) is significant as it exceeds the 5 per cent level of significance. The differences for Leunaphos IG No. 2 (11.364) and Nitrophoska IG No. 3 (16.950) are highly significant, surpassing the 1 per cent level of significance. The rank of the different treatments is also shown in table 3. Nitrophoska IG No. 3 ranked first; Leunaphos IG No. 2, second; Leunaphos IG No. 1, third; Superphosphate, fourth; and the control, fifth.

Table 4 shows the gain or loss per hectare under the different treatments. The computed average yield of shelled corn is also shown in the same table. Nitrophoska IG No. 3 gave the highest average yield (45.82 cavans) per hectare, followed by Leunaphos IG No. 2 (41.38 cavans), Leunaphos IG No. 1 (39.83 cavans), and Superphosphate (38.03 cavans). The control gave the lowest average yield, 31.33 cavans. The yield was computed on the basis of 58.5 kilograms per cavan, and the cost of one cavan was placed at ₱2.50. The increase in yield was highest in Nitrophoska IG No. 3 (14.49 cavans) and lowest in Superphosphate (6.70 cavans). The cost of the fer-

tilizer and its application was considered in determining the gain or loss. One ton (1,000 kilograms) of Superphosphate costs ₱75; Leunaphos IG No. 1, ₱170; Nitrophoska IG No. 3, ₱190; and Leunaphos IG No. 2, ₱155. The average time of application was 40.45 minutes per plot, or 37.5 hours per hectare. The cost of application was placed at ₱0.07 per hour, or ₱2.63 per hectare. The gain or loss was computed from the foregoing figures. All the different fertilizers showed gains. The highest gain, ₱14.60, per hectare was obtained from Nitrophoska IG No. 3 followed by Leunaphos IG No. 2 with ₱7.00, and Superphosphate with ₱6.62. The lowest gain was obtained from Leunaphos IG No. 1 with ₱1.62.

SUMMARY

1. The effects on the yield of the Native Yellow Flint variety of corn of Superphosphate, Leunaphos IG No. 1, Nitrophoska IG No. 3, and Leunaphos IG No. 2 were determined.

2. The average yield per hectare in cavans of shelled corn from the different treatments was as follows: control, 31.33 cavans; Superphosphate, 38.03 cavans; Leunaphos IG No. 1, 39.83 cavans; Leunaphos IG No. 2, 41.38 cavans; and Nitrophoska IG No. 3, 45.82 cavans.

3. The mean yield of each of the different fertilizers was higher than that of the control. Only the Superphosphate did not give a significant difference in yield over the control. Both Leunaphos IG No. 2 and Nitrophoska IG No. 3 gave highly significant differences, each exceeding the 1 per cent level of significance.

4. All the fertilizers showed gains. The highest gain per hectare was obtained from Nitrophoska IG No. 3, ₱14.60; and the lowest, from Leunaphos IG No. 1, ₱1.62.

LITERATURE CITED

MOTELLANO, P. L. 1916. A study of commercial fertilizers on corn. The Philippine Agriculturist and Forester 4: 217–230.

VIBAR, TORIBIO N. 1911. The influence of N-P-K on the growth and production of corn. The Philippine Agriculturist and Forester 1: 175–182.

TABLE 1

Yields in kilograms per plot of the different treatments in five blocks

TREATMENT	BLOCK					SUM	MEAN
	1	2	3	4	5		
Control	26.72	38.61	27.82	46.78	43.34	183.27	36.654
Superphosphate	31.64	42.76	46.85	52.18	49.05	222.48	44.496
Leunaphos IG No. 1	50.72	48.41	44.01	48.56	41.32	233.02	46.604
Leunaphos IG No. 2	51.80	48.43	49.06	54.24	38.56	242.09	48.418
Nitrophoska IG No. 3	59.00	48.41	48.79	59.28	52.54	268.02	53.604
Sum	219.88	226.62	216.53	261.04	224.81	1,148.88	
Mean	43.976	45.324	43.306	52.208	44.962		45.955

TABLE 2

Analysis of variance for yield in kilograms

SOURCE OF VARIATION	DEGREES OF FREEDOM	SUM OF SQUARES	MEAN SQUARE, OR VARIANCE	ESTIMATE OF VARIANCE [a]
Blocks	4	257.09	64.27	1.61
Treatments	4	768.16	192.04	4.82
Error	16	636.67	39.81	
Total	24	1,661.92		

[a] Values of F with 4 and 16 degrees of freedom are 3.01 and 4.77 for 5 and 1 per cent level of significance, respectively.

TABLE 3

Comparison of mean yield in kilograms of the different treatments [b]

TREATMENT	SUPERPHOSPHATE	LEUNAPHOS IG NO. 1	LEUNAPHOS IG NO. 2	NITROPHOSKA NO. 3	CONTROL	MEAN
Superphosphate	—	2.108	3.522	9.108	— 7.842	44.496
Leunaphos IG No. 1	— 2.108	—	1.414	7.000	— 9.950	46.604
Leunaphos IG No. 2	— 3.522	— 1.414	—	5.586	— 11.364	48.018
Nitrophoska IG No. 3	— 9.108	— 7.000	— 5.586		— 16.950	53.604
Control	7.842	9.950	11.364	16.950	—	36.654
Rank	IV	III	II	I	V	

[b] According to Fisher's "t" test of significance for 24 degrees of freedom based on critical difference, 8.21 and 11.13 for 5 and 1 per cent level of significance, respectively, were obtained.

TABLE 4

Gain or loss under the different treatments per hectare

TREATMENTS	RATE OF APPLICATION	AVERAGE YIELD OF SHELLED CORN	INCREASE IN YIELD OVER THE CONTROL	VALUE OF INCREASED YIELD	COST OF FERTILIZER AND APPLICATION	GAIN OR LOSS
	kilograms	*cavans*	*cavans*	*pesos*	*pesos*	*pesos*
Control	—	31.33	—	—		—
Superphosphate ...	100	38.03	6.70	16.75	10.13	6.62
Leunaphos IG No. 1	100	39.83	8.50	21.25	19.63	1.62
Leunaphos IG No. 2	100	41.38	10.05	25.13	18.13	7.00
Nitrophoska IG No. 3	100	45.82	14.49	36.23	21.63	14.60

BROOMCORN SEEDS AS A GRAIN FEED FOR LAYERS [1]

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Broomcorn (*Andropogon sorghum* Linn.) is grown only for the production of the brush used in the manufacture of brooms and brushes (Rothgeb, 1923). Morrison (1939) reported, however, that the seeds of broomcorn have considerable feeding value and may be saved by drying or ensiling. He added that in harvesting broomcorn the heads are cut before the seed has fully matured and the seed is removed from the brush before it is thoroughly dry.

In the manufacture of brooms and brushes, a good amount of seeds is produced as a by-product. A study was made to compare the value of broomcorn with yellow flint corn in a laying ration.

True (1900) states that broomcorn seed when allowed to ripen has considerable nutritive value; but that since it is necessary, in securing the best grade of brushes, to harvest the heads green, it has been found difficult to cure the seeds from them. He reports the proximate chemical analysis of broomcorn seeds as compared to corn kernels as follows:

SAMPLE	WATER	WATER-FREE MATERIAL				
		Ash	Protein	Fiber	N.F.E.	Fat
Broomcorn seeds ...	14.1	2.3	11.2	8.3	74.1	4.1
Corn kernels	10.9	1.7	11.7	2.4	78.1	6.1

From this analysis of the chemical composition of the ripe seed of broomcorn, he concluded that it is but slightly inferior to corn kernels as a food.

Morrison (1939) gives the following data on broomcorn seeds when used for animal feeding:

Total dry matter	88.6 per cent
Digestible protein	4.6 " "
Total digestible nutrients	55.7 " "
Nutritive ratio	1:11.0

[1] Experiment Station contribution No. 1444. Read before the Los Baños Biological Club, February 27, 1941.

Proximate chemical analysis:

Protein	10.8 per cent
Fat	3.5 " "
Fiber	8.4 " "
Nitrogen-free extract	62.7 " "
Mineral matter	3.2 " "

Digestible coefficients:

Protein	43.0 per cent
Fat	86.0 " "
Fiber	35.0 " "
Nitrogen-free extract	66.0 " "

PLAN OF THE STUDY

Forty-four six-month-old Los Baños Cantonese pullets, divided equally into two lots, were used in this study. The two lots were fed the following ration, all parts by weight:

LOTS	MASH			GRAIN	
	Fish meal	Copra meal	Rice bran	Corn	Broomcorn seeds
I (Corn)	20	20	60	100	0
II (Broomcorn)	20	20	60	0	100

Both lots were given the normal care, feeding, and management of the College flocks, the only difference being in the grain that they received. There was a preliminary feeding trial of one month to accustom the birds to the feed. In the broomcorn lot, the birds at first did not touch the grain part of their ration.

The birds were weighed at the end of each month. The feed consumption of each lot, mash and grain separately, was recorded for each week. Mortality records of each pen were kept. The birds in both lots were trapnested and a record of the eggs laid was kept for the duration of the experiment. Minor observations which were thought to bear on the results of the study were likewise noted.

The dried broomcorn seeds used in this experiment were obtained from the Farm Crops Division of the Department of Agronomy, a by-product of their manufacture of brooms from the said crop. The corn used was of the Laguna Yellow flint variety, grown by the Department of Animal Husbandry.

RESULTS AND DISCUSSION

Chemical composition. The proximate chemical composition of the two grains as found by analysis made by the Department of Agricultural Chemistry on an air-dry basis was as follows:

SAMPLES	BROOMCORN SEEDS	LAGUNA YELLOW FLINT CORN GRAINS
	per cent	*per cent*
Moisture	13.31	11.95
Fats (ether extract)	2.14	1.26
Ash	3.66	1.70
Proteins (N x 6.25)	4.13	10.44
Crude fiber	10.32	1.85
Carbohydrates (N.F.E.)	66.44	72.80
Calories per 100 grams	302.00	344.00

The broomcorn seeds used in this study are especially deficient in proteins, only 4.13 per cent, as compared to corn grains, 10.44 per cent. They are higher in fat and ash, and much higher in crude fiber, 10.32 per cent versus 1.85 per cent found in corn.

These results corroborate the data reported by Morrison (1939), especially with regard to the fiber content. The total digestible nutrients in flint corn is reported by the same author as higher, 84.1 per cent, than that of broomcorn seeds, 55.7 per cent; while the nutritive ratio of the former, 1:11.0, is wider than that of the latter, 1:10.4.

From these figures and from those reported by True (1900), it may be seen that although broomcorn seeds are comparable in a way to corn as a feed, still they are in general poorer in the quantity of nutrients they contain.

Body weights. In the allotment of the birds at the beginning of the experiment, care was taken to make the average body weights of the two lots as close as possible. Thus, both lots started with about the same body weight. The corn lot increased in weight faster than the broomcorn lot as the experiment progressed, but after the first half of the period, the broomcorn lot, in turn, became decidedly heavier during the latter half. The mean weight of the birds in the corn lot for the twelve months of the experiment was found to be 1.28 $\pm$ 0.020 kgm. while that of the broomcorn lot was 1.30 $\pm$ 0.022 kgm. The difference between these two means was found to be insignificant.

Feed consumption. In general the feed consumption of the corn lot birds was observed to be always lower than that of the broomcorn lot. The total feed consumption for the corn lot was 489.1 kgm., 228.4 kgm. of mash and 260.7 of grain. That of the broomcorn lot was 567.5 kgm., 305.3 kgm. of mash and 262.2 of grain.

The mean monthly feed consumption per bird was found to be as follows: Corn lot 0.90 ± 0.033 kgm. of mash and 1.03 ± 0.060 kgm. of grain; while that of the broomcorn lot was 1.27 ± 0.038 kgm. of mash and 1.08 ± 0.026 kgm. of grain. In the corn lot, the difference between the means of monthly grain and mash consumption per bird was insignificant. In the broomcorn lot, however, the difference of 0.19 ± 0.046 kgm. more mash than grain was significant. From these figures it can be deduced that the birds in the broomcorn lot did not relish the grain given to them as much as the corn lot birds relished theirs. Both the grain and the mash were given to the birds in quantities as much as they could consume with minimum of waste. The fact that the birds in the broomcorn lot consumed more mash than broomcorn seeds; when those in the corn lot consumed less mash than corn grain, shows the inferiority of broomcorn seeds to corn kernels as far as palatability is concerned. The analysis made by the Department of Agricultural Chemistry shows that broomcorn seeds have a much higher fiber content, 10.32 per cent, as compared to corn grains, 1.85 per cent. This fact may explain in part the low palatability of broomcorn seed when used as a grain feed for layers compared to corn grain.

During the month of July, both lots showed increased consumption of both grain and mash. It is interesting to note that the body weights of the birds in both lots in this month showed similar upward trend. At this time, it was noted that a majority of the birds in both lots had begun molting and putting on fat. According to Fronda (1928), the annual molt coincides in general with the gradual drop of egg production which begins about July and ends about December.

The average feed consumption per bird for a year was for the corn lot, and the broomcorn lot, 23.21, 28.14 kgms. respectively. These figures compare favorably with those observed by Fronda (1932) in the first Philippine egg laying contest of 27.43 kgm. for the Los Baños Cantonese pullets. However, they are quite low when compared with 30.04 kgm. for the same kind of birds observed by the same author in 1933 in the second Philippine egg laying contest.

Egg production. The average monthly egg production in per cent for both lots is given in table 1. Although the birds in both lots were of the same age, 6 months, at the start of the study, still the egg production for the first month differed greatly in favor of the corn lot, 36.36 per cent as against only 18.03 per cent for the

broomcorn lot. This is explained by the fact that not all the birds began laying at the same age. Whereas in the corn lot only four birds did not start laying until February, in the broomcorn lot nine birds did not.

From table 1, it can be seen that the distribution of egg production of the lots studied followed the general tendency of seasonal egg production. With the exception of one month, September, all throughout the duration of the experiment, the broomcorn lot had lower monthly egg production than the corn lot. Although the difference between the means of the two lots was not highly significant, (4.92 $\pm$ 1.859 per cent monthly egg production in favor of the corn lot) the consistently higher egg production of the corn lot is well apparent. The yearly per cent egg production of the two lots was as follows: corn lot, 32.80 and broomcorn lot, 27.82.

Protein is one of the essentials for egg production. Although both lots received equal amounts of animal protein supplement in the form of fish meal, analysis of the grains involved in the study shows that the broomcorn seed does not compare with corn as far as the protein content is concerned. Hence, this, in part may explain the lower egg production obtained in the broomcorn lot.

Mortality. Both lots started with twenty-two birds each. At the end of the experiment, only 16 birds were present in the broomcorn lot, and 19 in the corn lot. The first death occurred in the corn lot on July 31, the second on August 4, and the third on September 21. In the broomcorn lot, the deaths occurred on the following dates: May 27, July 23, August 1, August 23, November 2, and December 28. Post mortem examinations made on all birds that died showed no unusual fatal causes. Two birds from the broomcorn lot had to be killed because of an advanced stage of ocular roup. These occurred on August 1 and 23. The per cent of mortality of the birds in the two lots was found to be 13.64 for the corn lot and 27.27 for the broomcorn lot. Using the chi-square test (Snedecor, 1938), the difference in mortality of 13.63 per cent proved to be insignificant (Chi-square = 1.256).

Minor observations. The birds in the broomcorn lot generally molted earlier than those of the corn lot. At the end of the experiment there was a higher degree of pigmentation in the corn lot birds than in the other lot. This may be explained in part by the abundance of the carotenoid pigment called xanthophyll in the yel-

low flint corn fed to the layers. The presence of this pigment in broomcorn seeds is doubtful, and, if present, certainly not in abundance as it is in yellow corn.

Table 2 is a summary of the results obtained in this study.

SUMMARY

1. Broomcorn seeds are not as palatable as corn grains when fed to Los Baños Cantonese pullets.

2. Although the feed consumption of the broomcorn fed birds was higher than that of the corn-fed birds, the egg production was much higher in the latter lot than in the former, which shows that for egg production, broomcorn seeds are not as efficient as corn grains.

3. Where an abundant, cheap supply of broomcorn seeds may be had, however, broomcorn seeds may be used to advantage for feeding layers.

LITERATURE CITED

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TABLE 1

Average monthly egg production

MONTH	LOT 1 (CORN)	LOT 2 (BROOMCORN)
	per cent	*per cent*
January	36.36	18.03
February	42.86	39.45
March	43.25	36.80
April	38.48	34.85
May	33.28	27.73
June	34.24	27.78
July	31.96	26.91
August	27.08	25.60
September	22.33	24.07
October	23.77	21.68
November	26.14	22 89
December	30.56	25.57
Mean	32.53 ± 1.350	27.61 ± 1.247

TABLE 2

Summary of results

ITEMS	LOT 1 (CORN)	LOT 2 (BROOMCORN)
Number of birds at the start of the experiment	22	22
Number of birds at the close of the experiment	19	16
Mortality, per cent	13.64	27.27
Average weight per bird, kgm.	1.28 ± 0.020	1.30 ± 0.022
Average yearly mash consumption per bird, kgm.	10.84	15.14
Average yearly grain consumption per bird, kgm.	12.37	13.00
Average yearly feed consumption per bird, kgm.	23.21	28.14
Yearly egg production, per cent	32.80	27.82

COLLEGE AND ALUMNI NOTES

Luciano Valencia, junior, and Faustino Orillo, sophomore, were awarded the Bailon-Dela Rama scholarship in agriculture. They obtained the first two highest ratings in a competitive examination for the scholarship.

Mr. Ricardo T. Marfori '30, of the soil survey division, Department of Agriculture and Commerce, is leaving for the United States as a government pensionado. He will pursue graduate work in soil chemistry at the University of Illinois.

The faculty and students of the College are preparing for any emergency in accordance with decisions made at the faculty meeting on August 1. Lt. E. Reymundo has been giving lectures on volunteer guard work and Dr. R. Bersamin on first aid. A food committee was created to take charge of food supply in times of emergency. The students planted root crops.

Four Chinese visitors, Dr. Lin Kung-Hsiang of Lingnan University, Mr. Ting Su of Sun Yat Sen University, Dr. Ho Wen-chun and Mrs. Florence Pen Ho of West China Union University, were on the campus on August 25. They were on their way to China from America where they studied in various universities.

The following papers were read and discussed before the Los Baños Biological Club meeting on August 14:

Prof. F. B. Sarao. The Red Scindi in the College of Agriculture.

Mr. L. Villanueva and Mr. F. T. Lazaro. Lactic acid fermentation of sugar and final molasses by *Rhizopus oryzae* Went et Pr. Geerl.

Mr. D. R. Mendoza, Range of Philippine *Durio* spp.

President B. M. Gonzalez made an inspection of the College on August 14. He visited several departments and the Rural High School.

Dr. Leon G. Gonzalez, acting head of the Department of Agronomy, spoke before the National Research Council. He discussed his observations in connection with his trip to Java.

Mr. Rizalino Gilpo '41 is teaching exploratory and advanced courses in poultry and swine in Antique High School.

Mr. José Borromeo '36, assistant instructor in agricultural chemistry, and seventy-eight students of the College belonging to the reserve force of the Philippine Army were called to active military duty. Excepting six who were disqualified for reasons of physical defects, they form part of 20,000 Philippine Army reservists inducted on September 1 into the United States Army.

Professor José E. Velmonte, Acting Head of the Department of Agricultural Economics, resigned effective on September 1. He is now with the Bureau of Plant Industry.

Mr. Teofilo C. Briones '37 is now teaching at Nueva Vizcaya High School.

Mr. Mamerto E. Limuaco '24 was recently appointed teacher of agriculture in Laguna High School.

Mr. Francisco Saguiguit '40 was recently transferred from Odiongan Rural High School, Bohol, to Cavite High School.

Mr. Ruperto Denoga '34 is chief of the poultry division of Koronadal Valley Project of the National Land Settlement Administration.

THE EXPERIMENT STATION

LIST OF AVAILABLE CIRCULARS

Circular No. 2.—Bud Rot of Coconut (Revised, June, 1934) - - - *By G. O. Ocfemia*

Circular No. 3.—Experimental Errors and Application of the Probable Error to and the Interpretation of Experimental Results - - - - - - - - - - - - - *By Nemesio B. Mendiola* (Published as Chapter IV in *A Manual of Plant Breeding for the Tropics*, 1926, also sold by THE PHILIPPINE AGRICULTURIST at ₱3.25, paper bound, and ₱5.25, cloth bound, in the Philippines, and ₱3.50 and ₱5.50 elsewhere, postpaid.)

Circular No. 5.—Poultry Raising Made Easy (Revised) - - - - *By F. M. Fronda*

Circular No. 7.—How to Produce New Varieties of Gumamela (Hibiscus) - - - - - - - - - - - - *By Nemesio B. Mendiola*

Circular No. 8.—Horse Breeding in the Philippines - - - - - *By Valente Villegas*

Circular No. 9.—Fences for Farm Animals - *By B. M. Gonzalez and J. P. Esguerra*

Circular No. 10.—Practical Directions for Coffee Planting - - - *By Pedro A. David*

Circular No. 11.—The New College Copra Drier—Prepared in the *Department of Agricultural Chemistry* with the cooperation of the *Department of Agronomy and Extension*. (Revised by *Moises M. Kalaw*).

Circular No. 14.—Beriberi: Its Causes and Prevention - - - - - *By F. O. Santos*

Circular No. 15.—Cattle Raising under Philippine Conditions - - *By Valente Villegas*

Circular No. 16.—A Simple Farm Record for the Farmer - - *By Francisco M. Sacay*

Circular No. 17.—College Trapnest - - - - - - - *By F. M. Fronda and P. S. Paje*

Circular No. 18.—Surveying for Area with a Surveyor's Staff - *By Alexander Gordon*

Circular No. 19.—A Pageant: Illustrating the History and Growth of the College of Agriculture - - - - - - - - *By Anne F. Cole*

Circular No. 20.—Amount of Nutrients in Philippine Food Materials - - - - - - - - *By F. O. Santos and S. J. Ascalon*

Circular No. 22.—Goat Raising - - - - - - - - - - - - - *By Valente Villegas*

Circular No. 23.—Curing Pork and Making Sausage for Home Use - - - - - - - - - - - - - - *By Mariano Mondoñedo*

Circular No. 24.—Construction and Operation of Silos in the College of Agriculture - - - - - - - - - - - - *By José P. Esguerra*

Circular No. 26.—Training Cattle and Carabaos for Work - - - *By Valente Villegas*

Circular No. 27.—Bunchy-top of Abacá - - - - - - - - - - - *By G. O. Ocfemia*

Circular No. 28.—Cotton Culture - - - - - - - - - - - - *By Eulalio P. Baltazar*

Circular No. 29.—Collegiate Education in Agriculture - - - - *By L. B. Uichanco*

Circular No. 30.—What Should Filipino Ornamental Gardens and Ornamental Plants Be? - - - - - - - - - - *By N. B. Mendiola*

Circular No. 31.—How to Slaughter and Dress Farm Animals - - - - - - - *By Valente Villegas and M. Mondoñedo*

The price of the Circulars listed above is 10 centavos each except Nos. 5, 10, 16, and 30 which cost 20 centavos apiece.

ALL ORDERS SHOULD BE ADDRESSED TO THE BUSINESS MANAGER, THE PHILIPPINE AGRICULTURIST, AGRICULTURAL COLLEGE, LAGUNA, PHILIPPINES